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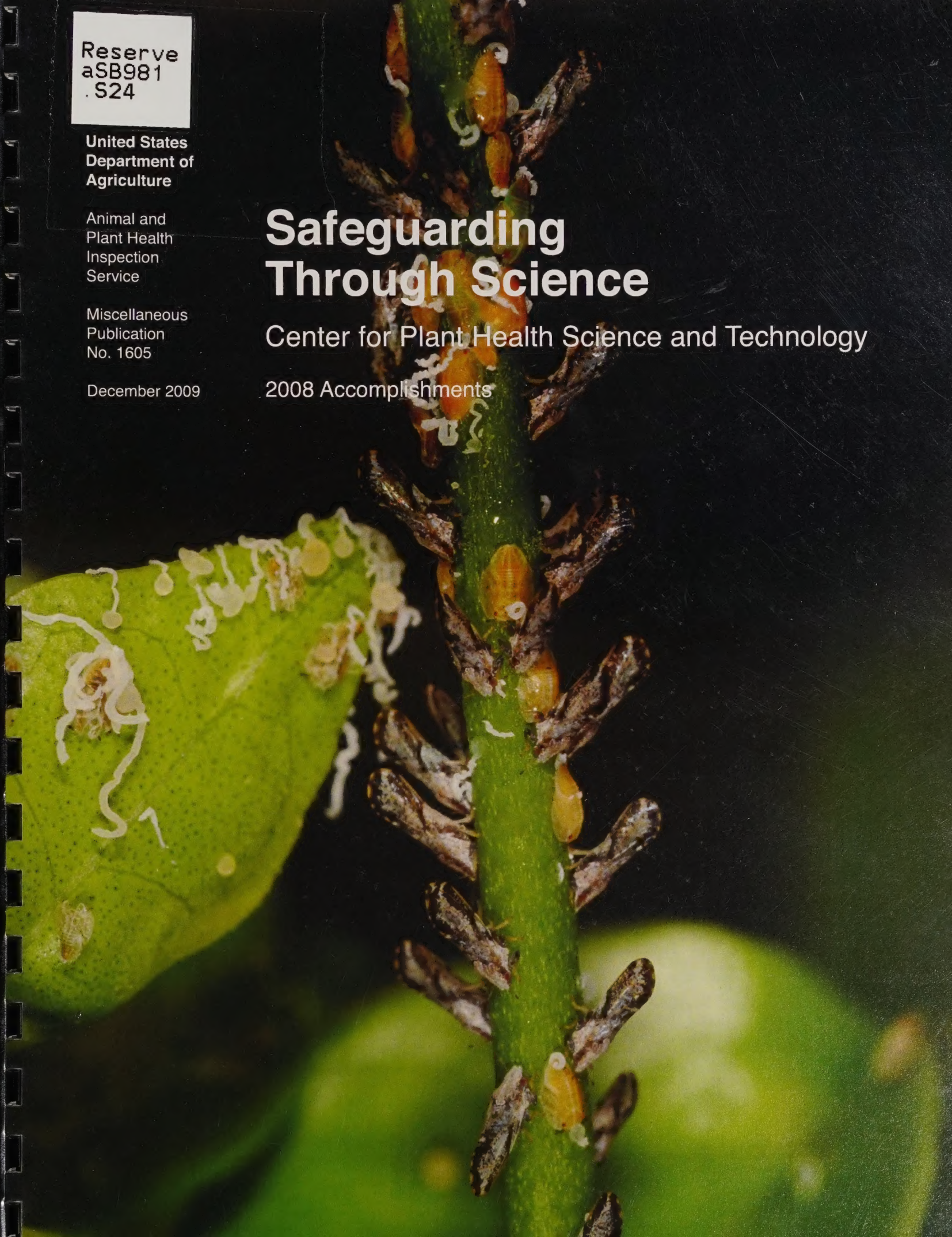
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Safeguarding Through Science

Center for Plant Health Science and Technology

2008 Accomplishments



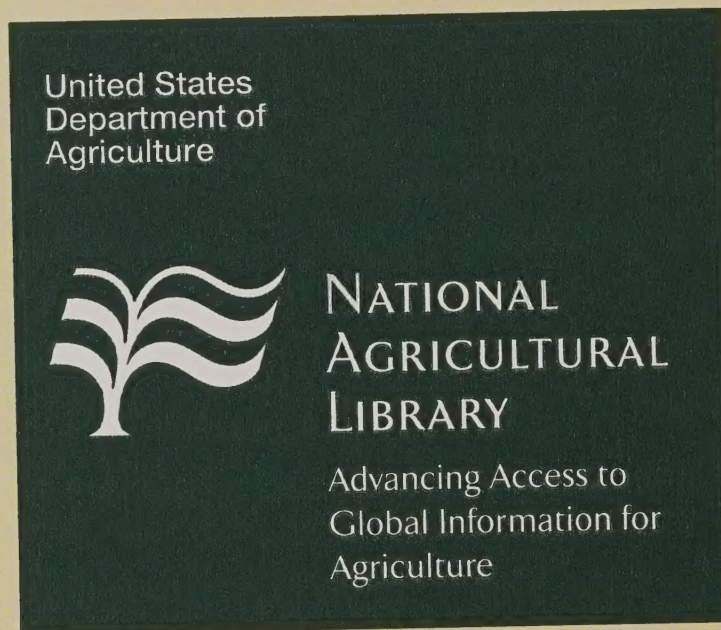
Center for Plant Health Science and Technology

Mission

The Center for Plant Health Science and Technology supports the regulatory decisions and operations of the Animal and Plant Health Inspection Service's (APHIS) Plant Protection and Quarantine (PPQ) program through methods development, scientific investigation, analyses, and technology.

Strategic Goals

- Enhance PPQ's efforts in pest detection and management
- Provide timely scientific and technical support required for emergency response and management
- Enhance support for APHIS trade-related plant health issues
- Provide current relevant scientific and technical information to PPQ decisionmakers
- Enhance PPQ's capacity to anticipate and respond to emerging scientific, technical, and regulatory issues through partnership



Cover photo: Asian citrus psyllid nymphs and adults on citrus (Photo provided by David Bartels, CPHST, Mission, TX).

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Message from the Director

The Center for Plant Health Science and Technology (CPHST) was designed and developed to support the regulatory decisions and operations of the Animal and Plant Health Inspection Service's (APHIS) Plant Protection and Quarantine (PPQ) program through methods development work, scientific investigation, analyses, and technology—all in an effort to safeguard U.S. agriculture and natural resources. While this is not a simple task, it is one to which all of us at CPHST are fully committed as we work to accomplish our mission. I lead and monitor this effort with the support and expertise of Bill Dickerson, the Associate Director for CPHST. This 2008 CPHST Annual Report is intended to offer an indepth look at the status of our programs and the progress we have made toward the Center's long-term strategic goals.

One of CPHST's most significant efforts in 2008 was to initiate efforts to improve the Center's organizational transparency and overall responsiveness to the needs of our stakeholders. As a result of our focus in this area, we are now developing a new workflow process that allows our customers to easily request and monitor projects and ensures that the highest priority projects are funded for successful delivery. This new system will allow us to more dynamically identify the needs of the agency, more effectively allocate and utilize resources, and provide our customers timely information regarding a project's status. Thus far, while still very much a work in progress, this new process is proving to be successful, and we will continue to advance and expand the service to our customers and staff.

In keeping with our efforts to anticipate stakeholder needs and improve our responsiveness, CPHST was involved in several emergency response programs in 2008. For example, we:

- Provided a survey plan for light brown apple moth (LBAM) in California and produced and supplied more than 200,000 lures for detection and delimitation surveys for the California Department of Food and Agriculture and the national LBAM survey;
- Established a laboratory colony of LBAM in Albany, CA, initiated the development of mass-rearing techniques, and supplied insects from the colony to researchers studying LBAM biological control; and
- Developed and conducted a demonstration project of the sterile insect technique for LBAM to manage and eradicate this aggressive plant pest (project still ongoing).

We continue to provide the best scientific support possible to our national and international partners to survey and control potato cyst nematode (PCN). Our lead scientists participated in technical working groups to review the PCN programs in New York and Idaho and helped to develop viability assay protocols that are essential for determining the efficacy of treatments. We have also worked with our neighbors in Canada to protect and secure both countries' potato industry. For example, we continue collaborating with the Canadian Food Inspection Agency to develop and implement PCN survey and detection guidelines to reduce unwanted pest introductions.

The considerable and growing concern of homeland security and the management of critical issues drives CPHST to lead the methods development of science-based systems for prevention, preparedness, response, and recovery. For example, we expanded national laboratory capacity in support

of national plant health emergencies by continuing development of the National Plant Protection Laboratory Accreditation Program and working closely with the National Plant Diagnostics Network (NPDN) labs to train 73 Federal, NPDN, and State diagnosticians in 2008. We continue to develop proficiency tests and deliver them to many laboratories and diagnosticians to ensure diagnostic capacity for U.S. agriculture.

We have also been very active in assisting the Citrus Health Response Program (CHRP). Many of our programs contribute to the CHRP by leading technical working groups, developing methods for area-wide control of the Asian citrus psyllid, and improving methods for citrus greening diagnostics and survey. We also provided valuable assistance to programs aimed at managing citrus canker, and we have several projects on other citrus pests.

These accomplishments are only a few of the examples of the scientific talent and excellence housed in CPHST and highlighted in this report. The organization is comprised of approximately 250 employees in 7 labs, 2 units, and multiple work stations in the United States, Colombia, and Guatemala. Our work is organized into five program areas: agricultural quarantine inspection and port technology, plant pathogens and weeds, response and recovery systems technology, trade issues and support, and domestic programs. Our scientists provide leadership and expertise in a wide range of fields—including risk assessments that support trade, domestic and offshore surveys and detection methods, geographic information systems, molecular diagnostics, biocontrol techniques, methods development for treatments, and mass rearing of insects. Moreover, CPHST scientists have provided critical and essential support for countless programs that address serious plant health threats, such as the Asian longhorned beetle, emerald ash borer, Mediterranean fruit fly (Medfly), Mexican fruit fly (Mexfly), Asian gypsy moth, *Sirex noctilio*, pink bollworm, sudden oak death (*Phytophthora ramorum*), plum pox virus, grasshopper/mormon cricket, and various invasive weeds.

CPHST's challenge for fiscal year 2009 will be to build on our strong base and focus on innovation. We continue to investigate, for example, new and/or improved diagnostic methods for arthropods and plant pathogens (including methods that may be applicable in ports or Plant Inspection Station environments), quarantine treatments, sampling and surveillance methods, and predictive modeling, to list only a few. There are other exciting projects already under development, including the new Agricultural Quarantine Inspection and Port Technologies Laboratory in Miami, FL, and numerous cutting-edge technology projects. We will continue to transition as our new Administration settles in and provides priorities and guidance.

CPHST is recognized nationally and internationally for its leadership in scientific developments to battle plant pests and diseases. We are proud of our accomplishments, and I hope you find this overview informative and educational.

Dr. Philip Berger, Director
Center for Plant Health Science and Technology
USDA, APHIS, PPQ

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National Science Programs

Agricultural Quarantine Inspection and Port Technology

National Science Program Leader—Dr. Mike Hennessey

Who We Are

The Agriculture Quarantine Inspection and Port Technology (AQI&PT) program provides scientific methods development support for agricultural inspections and treatment technologies to prevent the entry or spread of invasive quarantine pests. The AQI&PT program directly supports plant health officers and agriculture specialists stationed throughout the United States and at numerous international ports. APHIS-Plant Protection and Quarantine (PPQ) officers conduct treatments for commodities that need to meet import or export standards, inspect and certify commodities for export, and treat commodities for insects of regulatory concern. Agriculture specialists with the U.S. Department of Homeland Security's (DHS) Customs and Border Protection (CBP) inspect arriving international passengers and cargo for regulated agricultural commodities, foreign insects and diseases, and commodities that may be classified as biohazards. The AQI&PT program works to ensure that these APHIS-PPQ officers and CBP inspectors possess the most efficient, effective, and up-to-date treatments and equipment available for the inspection and treatment of our Nation's agricultural imports and exports.

Employees with the Treatment Quality Assurance Unit (TQAU), a branch within the AQI&PT program, provide primary support for AQI&PT activities. The AQI&PT program also works closely with USDA's Agriculture Research Service (ARS) and several Center for Plant Health Science and Technology (CPHST) laboratories across the country (Otis, MA; Mission, TX; and Fort Collins, CO), as well as foreign governments and stakeholders in private industry.

What We Do

The AQI&PT program's main function is to provide scientific support—both domestically and internationally—for PPQ and CBP inspection and treatment programs. AQI&PT also assists international agricultural organizations and private stakeholders in their work to minimize risks associated with invasive organisms. For example, AQI&PT reviews potential treatment methods to allow foreign commodities access into the U.S. market and evaluates treatment methods and equipment from domestic companies working to add a new treatment for a commodity.

In addition, the AQI&PT program works with ARS and foreign agricultural organizations to conduct scientific studies with an end goal of providing new and highly efficacious treatments for exotic pests. These treatments are then included in APHIS' *Treatment Manual*, an operational guide for PPQ and CBP inspectors that provides diverse and effective treatments for various pests and commodities.

In 2008, AQI methods development advanced in key areas due to the AQI&PT program's efforts. The program's most significant accomplishments from last year include:

- Improving packinghouse methods to reduce armored scale risk on Mexican avocados (fig. 1),
- Consulting on an AQI methods development center in Kenya,
- Developing a strategic plan for and sighting the new AQI Lab in Miami, FL,
- Completing a "Commodity Treatment Information System" that automatically tracks cold treatment on commodities in transit, and
- Serving as experts on the International Plant Protection Convention Technical Panel for Phytosanitary Treatments.



Figure 1. A washing and brushing technique to remove armored scales will be investigated on the packing line for Mexican "Hass" avocados (USDA-APHIS, CPHST photo).

Where We're Going

The AQI&PT program will continue supporting the daily work of agriculture inspectors and industry in the future. In 2009, the program's primary focus is to (1) establish and build a new AQI Lab at the port in Miami, (2) incorporate

hand-held gas chromatography technology into the Plant Inspection Stations to detect Rutaceae (fig. 2), and (3) develop a cold treatment for the Asian fruit fly (*Bactrocera invadens*).



Figure 2. Hand-held gas chromatograph (Z-nose) technology being used to determine if unknown plant sample is Rutaceae (citrus family) (CPHST photo).

Molecular Diagnostics and Biotechnology

National Science Program Leader—Vacant

Who We Are

The Molecular Diagnostics and Biotechnology (MDB) program provides scientific support to APHIS-PPQ programs for the characterization of pest organisms requiring molecular methods. The MDB program accomplishes this by developing and adapting appropriate technologies for molecular-based detection and identification of plant pests and pathogens, as well as by validating molecular assays for use by the regulatory community. The MDB program also supports the development of genetically modified (GM) organisms for use in pest management programs. In addition, the program is involved in the development of the USDA National Plant Protection Laboratory Accreditation Program (NPPLAP), where diagnostic determinations requiring a regulatory response can be performed by governmental and affiliated nongovernmental laboratories outside PPQ and be recognized as valid.

The MDB program's vision is to identify and employ effective molecular-based technologies to ensure that APHIS delivers successful plant health programs. To reach this goal, these assays must be highly reproducible, robust, and responsive to program needs. With this in mind, the MDB program:

- Evaluates and develops new technologies for molecular diagnosis of exotic pests and pathogens,
- Validates molecular diagnostics,
- Develops and deploys proficiency tests for external laboratories,
- Trains collaborators on the use of specific molecular diagnostics,
- Develops quality assurance for accepted, validated molecular diagnostics, and
- Supports the environmental assessment of transgenic insects for use in regulatory programs.

What We Do

The MDB program provides science-based support for several PPQ programmatic efforts, such as the Citrus Health Response Program (CHRP) and the *Phytophthora ramorum* (sudden oak death [SOD]) program. MDB also supports improved responses to emerging pest introductions in the United States, including the recent potato cyst nematode (PCN), *Globodera pallida*, and plum pox virus (PPV) finds. In 2008, the MDB program sponsored technical working groups to provide science-based recommendations when new research findings or changes in these pest situations occurred.

The MDB program also continued to facilitate the further development of the NPPLAP, with the goal of increasing APHIS-PPQ's capacity to meet potential emergency diagnostic needs using standardized methods. This work focused on diagnostic protocol standardization and laboratory proficiency testing for molecular diagnosis of high-consequence pests.

In 2008, the MDB program continued to support the Integrated Consortium of Laboratory Networks (ICLN), which includes 10 Federal agencies or departments that have agreed to work cooperatively in optimizing national laboratory preparedness and improving the coordination of laboratory response. The ICLN is a DHS infrastructure with a coordinated and operational system of laboratory networks that provide timely, high-quality, and interpretable results for early detection and effective consequence management of acts of terrorism and other events requiring an integrated laboratory response. The MDB staff belongs to the ICLN Network Coordinating Group, as well as a variety of subgroups addressing training, proficiency testing, laboratory accreditation and quality assurance, methods development, scenario

development, and information technology. In 2008, the ICLN developed its charter as well as a “Response Architecture” document that provides a template for the coordination of laboratory networks to provide diagnostic capacity as a nationwide emergency response.

The MDB program also continued to assess emerging technologies for their potential to improve APHIS-PPQ program management (i.e., by increasing capacity or streamlining current efforts). In addition, the MDB program provided guidance to decisionmakers on how new technologies could impact the safeguarding of U.S. agriculture from accidental or deliberate introductions of high-consequence pests.

Where We’re Going

The use of molecular methods within APHIS is rapidly increasing to meet today’s plant health challenges. The need for MDB’s services will only increase further as our society continues to face new challenges in safeguarding U.S. agriculture and natural resources due to increased trade, faster delivery times, and new commodities. As the primary support for molecular diagnostic methods within APHIS-PPQ, MDB personnel will continue to look into emergent molecular technologies for potential adoption by the agency’s plant health safeguarding programs. We will also keep the developers of these new technologies informed about PPQ’s unique needs so that future technologies are developed with these needs addressed.

Through our involvement with the NPPLAP and other activities, MDB will also be expanding on efforts to engage and coordinate the molecular diagnostic capacity outside the APHIS-PPQ system. The continued development of accreditation and certification programs will increase the quality of procedures and reporting expectations at participating laboratories systemwide. These programs are designed to produce protocols that are at the forefront of diagnostic capacity and adopted throughout the United States and internationally as the standards for molecular protocols and diagnosis.

Risk and Pathway Analysis

National Science Program Leader—Dr. Ron Sequeira

Who We Are

The Risk and Pathway Analysis (RPA) program focuses on collecting and interpreting scientific evidence and technical information regarding plant pest risks. RPA products help APHIS-PPQ design risk-based policies and regulations for import, export, and domestic programs. The work of the RPA staff is essential to identify and assess new pest threats (fig. 3), provide scientific support for regulatory updates and revisions, and help prioritize resources to maximize plant health safeguarding capabilities.

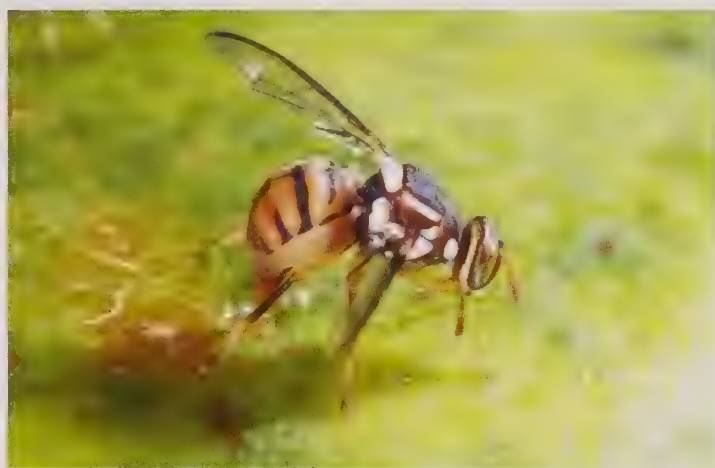


Figure 3. The Oriental fruit fly (*Bactrocera dorsalis*), a polyphagous destructive pest that can cause severe economic losses (Photo by Scott Bauer, USDA-ARS).

RPA is supported primarily by CPHST’s Plant Epidemiology and Risk Analysis Laboratory (PERAL) in Raleigh, NC, with satellite staff in Hawaii and Bogotá, Colombia. PERAL collaborates with other CPHST laboratories, other groups within APHIS-PPQ and USDA (e.g., ARS), academia, foreign counterparts, and stakeholders in developing scientific analyses that are essential for the formulation, implementation, and modification of risk-based phytosanitary policies and procedures. RPA program capabilities have expanded rapidly over the past few years to become a central function for PPQ and an international benchmark for pest risk analysis, or PRA.

What We Do

The foundation of RPA's analytical capacity is its diverse, high-caliber scientific staff—which includes entomologists, plant pathologists, botanists, ecologists, economists, and other specialists—along with its extensive, unique, and ever-growing collection of information on plant pests. RPA uses state-of-the-art tools and methodologies for pest risk assessment, including sophisticated spatial technology systems that integrate weather, pest distribution, and other databases to analyze pest dynamics, identify agro-environmental pests of greatest concern, and identify potential pathways for the introduction of harmful exotic pests and predict their spread.

In 2008, the RPA program led APHIS in the development of analytical products to support emergency, domestic, and trade programs. Specific, high-visibility, and controversial risk and pathway analysis products included those for light brown apple moth, citrus canker, citrus greening, and solid wood packing materials. In addition, the RPA program addressed high-profile trade issues linked to commodity importations. Several RPA initiatives responded to White House and other executive mandates. High-profile commodity risk analyses included those supporting free trade agreements and those deemed significant by the Office of Management and Budget. The RPA program processed more than 24 commodity import requests in 2008.

Where We're Going

RPA's future plans include promoting the continued evolution of risk analysis tools and methodologies while also increasing the transparency and efficiency of risk analysis work processes. We also expect to give increasing emphasis to risk assessments for pest plants (noxious weeds) and propagative material. These plans are directed to continue helping APHIS-PPQ design risk-based policies and regulations based on scientific and technical information for import, export, and domestic programs.

Response and Recovery Systems Technology

National Science Program Leaders—Dr. Russ Bulluck (pathogens) and Mike Stefan (arthropods)

Who We Are

The Response and Recovery Systems Technology (RRST) program provides timely scientific support to regulatory program managers and decisionmakers when high-consequence

plant pests have been detected in the United States. The program emphasizes preventive measures that minimize the chances that introductions will occur and develops rapid response and recovery technologies geared towards eradicating or containing newly detected pests.

What We Do

RRST supports a wide range of plant health programs, which are described in more detail below. As with all PPQ work, these programs use integrated pest management strategies, phytosanitary measures, and technologies to combat exotic diseases and pests while minimizing adverse effects on the environment, producers, and consumers.

Citrus Health Response Program

The CHRP was formed in response to the establishment of citrus canker (fig. 4) and citrus greening (also known as huanglongbing [HLB]) in Florida and the potential for other pests and pathogens to enter U.S. citrus-production areas. The objective was to develop a science-based approach to manage citrus canker. The program is based on citrus disease management using harmonized production standards across all citrus-producing States. A technical working group (TWG) was convened to review, define, and identify conditions under which nursery stock intended for shipment to non-citrus-producing States can be produced and shipped with no (or very low) risk of movement of citrus canker, HLB, or Asian citrus psyllid (the vector of HLB).



Figure 4. Citrus canker symptoms on grapefruit (Photo by Esther Serrano, APHIS-PPQ).

Light Brown Apple Moth

The light brown apple moth (LBAM), *Epiphyas postvittana*, (fig. 5) was detected and confirmed in Alameda County, CA, in March 2007. If left uncontrolled, LBAM could cause significant damage to many different crops, including stone fruits, pome fruits, grapes, and citrus. A TWG comprised of scientists from the United States, Australia, and New Zealand was formed to make recommendations for potential eradication or control of the pest, develop management recommendations, assess research needs, and provide ongoing scientific input to program operations. In 2008, CPHST initiated and coordinated activities based on the pest management recommendations and research needs identified by the TWG. Currently, the program's primary focus is to develop sterile insect technology for the control of LBAM.



Figure 5. Light brown apple moth (Photo by Todd Gilligan, Colorado State University).

Panicle Rice Mite

Since its confirmed detection in July 2007, efforts to eradicate the panicle rice mite (PRM), *Steneotarsonemus spinki*, have continued at rice research facilities—including fields and greenhouses—in the United States and Puerto Rico. A TWG of mite and rice experts was assembled to develop management recommendations for this serious rice pest and advise the program on the feasibility of eradication efforts. In 2008, RRST helped develop treatments to facilitate the movement of rice seed from Puerto Rico. The program will continue to provide scientific support as additional research facilities are identified and regulatory actions are taken.

Fruit Fly Introductions

The RRST program provides scientific support to eradication efforts for Mediterranean fruit fly (Medfly), *Ceratitis capitata*, and Mexican fruit fly (Mexfly), *Anastrepha ludens*, outbreaks

in California, the Mexfly eradication program in Texas, and the Moscard barrier program in Guatemala. In 2008, efforts to gather and analyze ecological data on Medfly resulted in the development of more effective areawide management tactics for use in the Guatemala Moscard barrier program's coffee belt. Pilot studies were also ongoing for the development of a much-needed bait station technology for use during Medfly outbreaks and as part of the Moscard barrier program. The RRST program also initiated genetic barcoding studies to support the source identification of Oriental fruit fly and other *Bactrocera* spp. outbreaks.

Red Palm Mite

Red palm mite (RPM), *Raoiella indica*, (fig. 6)—a pest of coconuts and ornamental palms in Asia and Africa—arrived in the Caribbean in 2004. By 2008, the pest had spread to most islands, including Puerto Rico and the U.S. Virgin Islands, and portions of Florida. RPM has already had serious impacts on the region's coconut and banana industries and is expected to cause economic damage to tropical and subtropical agriculture wherever palms are grown in the Western Hemisphere. CPHST chairs the Red Palm Mite TWG, which was formed after discovery of the pest to develop management recommendations, support continued surveillance activities, and oversee the assessment of biological control options for this important pest.



Figure 6. Red palm mite (Mounting and collecting was done by R. Ochoa & E. Kane) (Photo by E. Erbe, USDA-ARS).

Plum Pox Virus

Several PPV science panels were convened in response to recent detections of the virus in New York, Michigan, and Canada. One of the panels' tasks was to review the current eradication program in Pennsylvania and outbreaks in New York and Michigan in order to analyze the program components that may be applicable to newer finds and situations. Recommendations from the science panels were used to focus and prioritize program components suitable for phytosanitary actions.

Potato Cyst Nematodes

In response to a 2006 detection of the PCN in Idaho potato fields, several TWG meetings for pale cyst nematode, *Globodera pallida*, and the golden nematode, *Globodera rostochiensis*, were convened to discuss the pest's status in North America in vital potato production areas—including Alberta, Idaho, New York, and Quebec—and to make management recommendations based on the most current scientific knowledge and expertise. These TWGs were charged with providing scientific support to the Idaho and New York regulatory programs based on timely information as the response infrastructure developed. In addition, in a national survey for PCN, the TWGs provided support on issues related to the survey and detection of the pest.

Phytophthora ramorum

CPHST provided ongoing scientific support for the *P. ramorum* program by participating in science panel meetings, developing and implementing TWGs, and recommending modifications to eradication protocols as new scientific information became available. Rapid incorporation of new scientific developments continued to be a major driver of our response and recovery efforts for *P. ramorum*, the SOD pathogen. CPHST also continued to be responsible for the development of the National Ornamentals Research Facility at Dominican University of California. Activities at this facility will provide critical information on the fate of *P. ramorum* in nursery locations with a focus on developing methods to effectively mitigate the pathogen.

Gladiolus Rust

TWG meetings were held in 2008 to review information on the biology of the gladiolus rust pathogen (*Uromyces transversalis*) (fig. 7), evaluate the control recommendations already in practice for this disease, and assess progress towards eradication. These meetings focused particularly on Florida's efforts in eradication. An additional TWG meeting to re-

evaluate the feasibility of eradication, management options if eradication is deemed infeasible, and ongoing research needs to support regulatory decisionmaking has been planned for 2009 and will include both Florida and California.

Where We're Going

The RRST program continues to evolve as it seeks to meet the challenges presented by an increasing arrival rate of exotic diseases and pest species. RRST also continues to contribute to the development of and participation in training exercises related to plant health emergencies. We will continue to expand our abilities to provide scientific support to plant health emergencies with a focus on providing information that is scientifically relevant to the particular pest and operationally practical. We will also contribute to the preparation and development of New Pest Response Guidelines with APHIS-PPQ's Emergency and Domestic Programs. RRST will focus on using the output from pathway analyses and overseas information-gathering efforts to prepare for future pest threats and support the development of new management programs.



Figure 7. Rust pustules commonly observed with gladiolus rust (APHIS file photo).

Survey Detection and Identification

National Science Program Leader—Dan Fieselmann

Who We Are

The Survey Detection and Identification (SDI) program provides sound science and demonstrated methodologies to PPQ for rapid detection and identification of exotic pests.

The SDI program is divided into four cross-functional groups: early detection, identification technology, spatial technology, and biological control. SDI supports PPQ programs through ad hoc and long-term projects, committee memberships, and virtual teams. Additional support is gleaned through cooperation with academic institutions, industry, and government agencies.

The SDI early detection team supports national pest surveillance through the PPQ Cooperative Agricultural Pest Survey (CAPS) Program. SDI provides survey support to CAPS through pest prioritization, commodity-based survey guidelines, trap and lure development, and offshore methods development (fig. 8). In addition, SDI provides support to emergency programs through survey development.



Figure 8. As part of the Panama Canal Zone Exotic Mollusk Survey Project, a technical training session was held at the University of Panama and the Panama Canal Zone area to train participants from the University, Sanidad Vegetal, and Cuarentena on how to survey for, identify, and chemically control invasive snails and slugs (Photo by Amy Rhoda, CPHST, Miami, FL).

The SDI identification technology team develops Lucid taxonomic tools (fig. 9) and provides training and technical support for Lucid tools and other computer-based identification resources. Additionally, the group has developed an automated system for sorting and identifying insects using camera recognition technology and robotics.

The SDI spatial technology team provides support in the fields of geographic information systems (GIS) and remote sensing. Examples include risk maps in support of CAPS, national, and emergency programs (fig. 10); Web-based spread modeling; and remote sensing to detect the efficacy of weed control efforts.

The SDI biological control team develops and matures technology to eradicate or manage exotic organisms. The process includes identifying natural enemies and competitors, developing rearing technologies and release techniques, and monitoring released biological control agent populations.

What We Do

The SDI program provides scientific support for the CAPS program, which includes developing commodity-based surveys and risk maps. Commodity surveys focus on economically important crops and provide an efficient method of surveying for many exotic pests within a specific crop. In 2008, SDI delivered three commodity survey reference documents and survey guidelines for grape, small grains, and pine to the CAPS program for field use. Risk maps assist CAPS cooperators in survey planning and resource allocation. In 2008, the SDI spatial technology team developed and improved risk maps for pests on the CAPS Prioritized Pest List. National maps were generated at the county scale using host data, biological data, and a combination of the host and biological data. The CAPS Pest Risk maps can be accessed at www.nappfast.org.

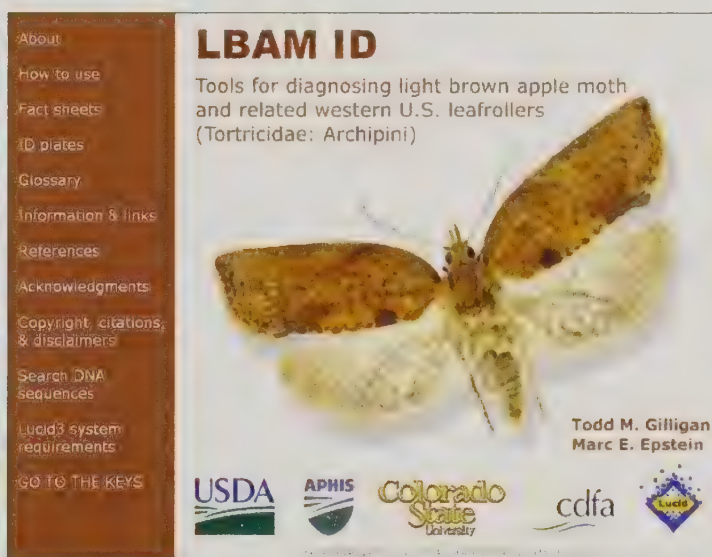


Figure 9. CPHST, in cooperation with Colorado State University and the California Department of Food and Agriculture, is creating an Internet-based identification tool—LBAMid—for light brown apple moth and related western U.S. leafrollers to aid survey and detection efforts (Image by Terrence Walters, CPHST, Fort Collins, CO).

The SDI program develops novel technologies for the detection and identification of pests. In 2008, through a cooperative agreement with Pennsylvania State University, SDI developed a working prototype of a new type of insect trap. The trap uses acoustic technology to pick up a signal from a particular insect wing beat and notify the home base remotely.

One of SDI's strategic objectives is to provide ongoing learning opportunities to individuals interested in developing and using Lucid tools to support PPQ's survey, detection, and identification needs. In 2008, SDI's identification technology team conducted five domestic workshops and provided materials and electronic keys for two international identification workshops. SDI continued to develop Lucid identification tools for a variety of government users to enhance and complement existing paper-based and electronic identification resources. In addition, SDI delivered four Lucid interactive Internet-based tools to APHIS and its domestic and international cooperators.

The SDI program also identifies natural enemies for pests of concern and develops rearing and release methods for their implementation in pest control efforts. In 2008, offshore research included studies to identify natural enemies of the coffee mealybug, evaluate the impact of local natural enemies of red palm mite, and determine the efficacy of two passion-vine mealybug parasitoids that were discovered controlling the pest in Trinidad. In addition, SDI conducted field trials on natural enemies of emerald ash borer, tropical soda apple, and Asian citrus psyllid.

Where We're Going

Decisionmakers recognize the urgent need for early detection of plant pests. Tremendous plant resources are vulnerable to pest introductions. Early detection of pests often provides significant long-term savings by reducing pest control costs and increasing the feasibility of eradication. In response, leaders have increased pest detection resources. The SDI program will continue to develop and improve tools and technologies that promote early detection and identification of exotic pests.

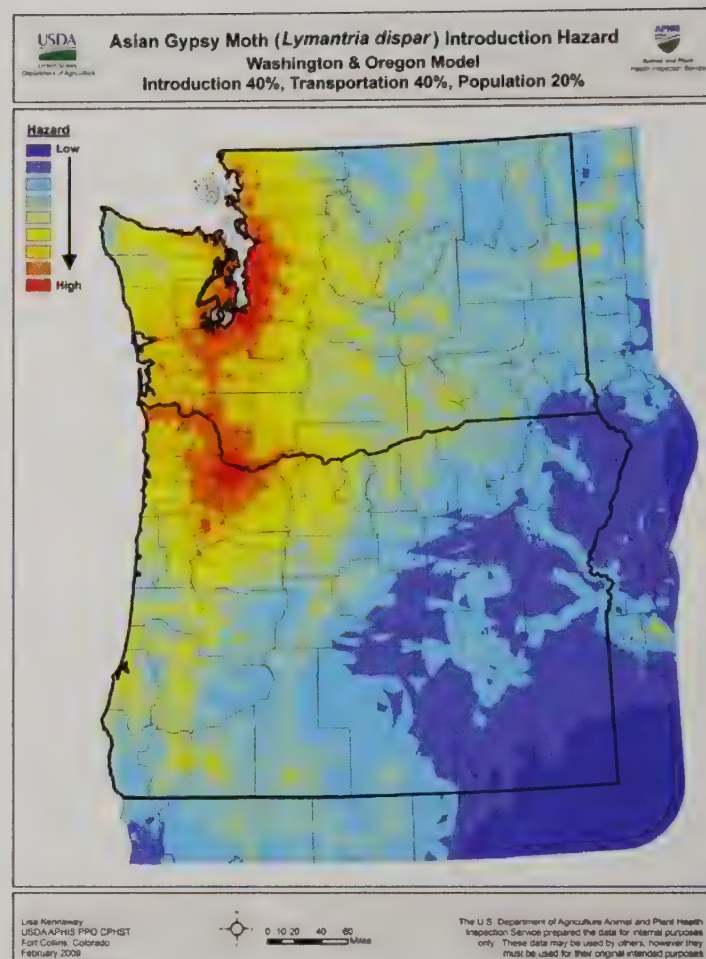


Figure 10. CPHST, in cooperation with the Washington and Oregon State departments of agriculture, is developing a spatial model that predicts areas with highest potential for Asian gypsy moth introduction (Image by Lisa Kennaway, CPHST, Fort Collins, CO).

A photograph of a large, green plant, likely a species of giant hogweed, with several large, flat-topped clusters of small white flowers (umbels) against a dark background. The plant has large, deeply lobed leaves and thick, green stems. The text "Laboratories and Units" is overlaid in white serif font on the right side of the image.

Laboratories and Units

The Center for Plant Health Science and Technology (CPHST) is comprised of seven laboratories and two units (fig. 11). A new laboratory in Miami, FL, is under development and will open in 2009.

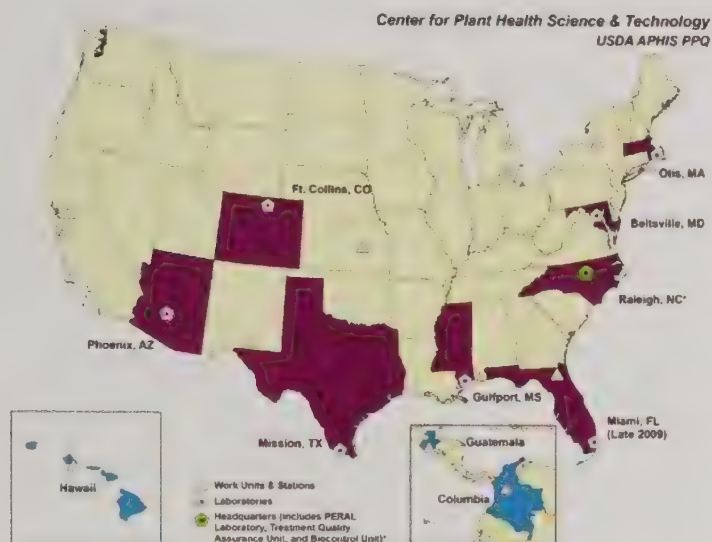


Figure 11. CPHST lab, stations, and unit locations (Image by Lisa Kennaway, CPHST, Fort Collins, CO).

CPHST Lab, Beltsville, MD

Contact—Dr. Laurene Levy

How We Support the Mission

The CPHST Beltsville Lab investigates cutting-edge methods and technology from the fields of plant pathology, molecular biology, human and animal clinical diagnostics, and biodefense to develop, adapt, and improve methods for accurate and rapid diagnosis of plant pathogens. In collaboration with our scientific partners, we focus applied research for USDA laboratories, land grant universities, international research centers, and other external sources such as national and U.S. military laboratories to further USDA regulatory programs. Our scientists validate and clarify biochemical and molecular diagnostics for stakeholder release and develop standard operating procedures with clearly defined diagnostic instructions.

We also achieve timely delivery of progressive technology that is field-deployable and uncomplicated in its operation primarily for Plant Protection and Quarantine (PPQ) emergency response and eradication programs. For example, in 2008, we developed methods for plant pathogen detection in foreign germplasm for surveys and at Plant Inspection Stations. The Beltsville Lab also uses advanced diagnostic meth-

ods for accurate and rapid detection of high-consequence and U.S. Select-Agent plant pathogens to fulfill Federal confirmatory testing requirements when operational methods are not yet available (e.g., during first U.S. detections or nonroutine scenarios).

As a key component of the CPHST National Plant Pathogen Laboratory Accreditation Program (NPPLAP), the Beltsville Lab is responsible for Proficiency Test (PT) panel development, panel deployment, and evaluation of PT results. The lab also provides preparedness and outreach to the plant pathology diagnostic community by supplying standard operating procedures and hands-on diagnostic laboratory training to scientists from the National Plant Pathogen Diagnostic Network (NPDN), land grant universities, PPQ (including the Plant Health Program's Molecular Diagnostics Laboratory), State departments of agriculture, and international regulatory programs.

Recent Accomplishments

- Conducted a comparison of five published, real-time polymerase chain reaction (PCR) assays for the detection of *Phytophthora ramorum*, the sudden oak death (SOD) pathogen. Based on the comparison, the lab released a validated, real-time PCR assay based on the elicitin gene for use with the current real-time internal transcribed spacer (ITS) PCR assay. The new detection system, a combination of the ITS and elicitin real-time PCR assays, increases the speed and capacity of labs performing SOD diagnostics.
- Developed a real-time PCR assay for detection of the *Candidatus Liberibacter* spp. associated with the zebra chip disease of potato.
- Developed a simplified one-tube, real-time PCR assay for the specific detection of high-consequence potato cyst as well as the closely related tobacco cyst nematodes (*Globodera pallida*, *G. rostochiensis*, and *G. tabacum*).
- Developed a microelectronic DNA chip assay to detect and differentiate plum pox virus strains. We also developed a DNA chip assay to detect and differentiate *Globodera* species, including *G. pallida*, *G. tabacum*, and *G. rostochiensis*.
- Validated and deployed a tissue-based proficiency test (PT) panel to NPPLAP in the fourth year of the *P. ramorum* certification program. We also developed and validated a citrus tissue-based PT panel for labs seeking

certification for citrus greening (also known as huanglongbing [HLB]) detection. The citrus greening PT panel is the first U.S. select-agent PT panel developed and distributed by the NPPLAP.

- Conducted hands-on laboratory training for the detection of citrus greening (fig. 12), potato cyst nematode, potato brown rot, citrus canker, SOD, and plum pox to increase the preparedness of over 70 scientists from PPQ, the NPDN, land grant universities, State departments of agriculture, and international regulatory programs. HLB training was also provided to ten Secretaría de Agricultura, Ganadería, Desarrollo Rural, Pesca y Alimentación (SAGARPA) phytosanitary scientists onsite in Queretaro, Mexico.
- Served on Integrated Consortium of Laboratory Networks (ICLN) sub-committees for methods development, accreditation and quality control, proficiency testing, and training. The ICLN is a consortium of 10 Government agencies that promotes common standards among all Government diagnostic networks and improves the coordination of laboratory responses to incidents. Beltsville Lab scientists contribute to PPQ task forces, participate in working groups to develop recovery plans for the National Plant Disease Recovery System, and serve as subject experts for professional journals and on panels for U.S. and international scientific grant agencies.



Figure 12. Blotchy mottle symptoms of HLB on lime (Photo by H. Gomez, PPQ).

CPHST Lab, Fort Collins, CO

Contact—Dr. Richard Zink

How We Support the Mission

The Fort Collins Lab develops science-based methods, innovative tools, and state-of-the-art technology to reduce levels of risk associated with new and established weed species and imported plants for planting, and transfers these tools to PPQ and other Federal and State agencies. In addition, the Fort Collins Lab provides scientific support for pest detection and survey methods under the Cooperative Agricultural Pest Survey (CAPS) Program. The laboratory transfers methodologies and tools to field operations, ensuring efficient and effective survey, detection, identification, emergency response, and eradication. As the primary source for new technologies, the lab develops electronic, matrix-based identification resources to support rapid, consistent, and accurate identification and nomenclature of pest species.

The Fort Collins Lab is typically APHIS' clearinghouse for the management of invasive plants and plants on the Federal Noxious Weeds list through applications of biological, chemical, and/or cultural controls. PPQ operational programs receive spatial technology support from the Fort Collins Lab to guide them in the application of new geospatial survey and detection methods. At a satellite lab in Albany, CA, staff members develop artificial diets and rearing systems for light brown apple moth (LBAM), *Epiphyas postvittana*, and biocontrol agents. From a second satellite lab in Las Cruces, NM, personnel provide engineering expertise in the automated screening of biological materials for detection of alien species posing potential risks to agriculture and natural resources.

Recent Accomplishments

- Established a breeding colony of LBAM to support mass rearing research, molecular diagnostics, and biological control initiatives.
- Supported the CAPS community by developing grape commodity-based survey guidelines, updating and reformatting the soybean commodity-based survey guidelines, and establishing the protocol for writers of commodity-based survey references. Initiated the New Pest Response Guideline for exotic *Phytophthora* spp. in nurseries and forests.

- Developed a spatial model to predict areas with the highest Asian gypsy moth (AGM), *Lymantria dispar*, introduction potential based on transportation and population variables.
- Through a cooperative project with Kansas State University, developed an interactive pest spread model to help analyze the movement of a pest or disease based on host availability. The model is intended for APHIS risk assessors and emergency responders as well as State cooperators trapping for pests with the CAPS program.
- Provided Lucid training to PPQ's Professional Development Center. We also delivered Lucid identification tools for "Invasive Ants of the Pacific Basin," "Wood Boring Beetles of the World," "Pests and Diseases of Small Grains," and "Common Grasshoppers of the Western United States" (Editions 2.0 and 3.0). In addition, we developed a Lucid tool for LBAM and easily confused taxa and presented two, 2-day workshops for PPQ and cooperators on the development of Lucid tools.
- Confirmed hyperspectral imaging of taxonomically significant spectral patterns (from ultraviolet to infrared) within and among pest taxa for counting, sorting, and classifying trap sample specimens.
- Demonstrated practical levels of suppression of two highly invasive plants—onionweed (*Asphodelus fistulosus*) (fig. 13) and benghal dayflower (*Commelina benghalensis*)—with novel combinations of herbicide rates, spray additives, application timing, and cover crops.



Figure 13. Onionweed in flower (Photo by Craig Ramsey, CPHST, Fort Collins, CO).

CPHST Lab, Gulfport, MS

Contact—Anne-Marie Callcott

How We Support the Mission

The Gulfport Lab consists of an Analytical Chemistry section and an Imported Fire Ant (IFA) section. The Analytical Chemistry section conducts routine sample analysis for detecting the presence of pesticide residues and toxic substances, which directly supports ongoing APHIS operational and emergency programs, such as IFA, Asian longhorned beetle (ALB), boll weevil, grasshopper, and fruit fly. In addition, the section supports APHIS projects by providing chemistry-based options for PPQ field operatives concerning the identification and detection of prohibited commodities as well as the detection of invasive insect species. The IFA section develops methods and tools for the survey, detection, regulation, and chemical and biological control of the imported fire ant (*Solenopsis invicta*) (fig. 14). Technology developed by the IFA section is utilized by PPQ, State Plant Regulatory Officials, the nursery industry, the chemical industry, farmers, homeowners, and other stakeholders.



Figure 14. Imported fire ant (APHIS file photo).

Recent Accomplishments

- Analyzed 1,050 APHIS routine program support samples and reported within stipulated deliverable times.
- Completed 1,100 project samples supporting APHIS initiatives.
- Analyzed soil samples from APHIS' International Services (IS) in Suriname, South America, for elemental tin using inductively coupled plasma mass spectrometry analysis.

- Developed an analytical method for detecting imidacloprid in honey, which supports limited sampling of regional honey and allows us to verify that ALB treatments do not impact this important commodity. The method utilizes a complex liquid/liquid extraction and concentration preparation coupled with high-pressure liquid chromatography analysis.
- In support of USDA's initiative to reduce costs and waste associated with chemistry applications, we modified methods for routine analytical processes that support programs, such as IFA and ALB, resulting in an approximate 40-percent reduction in both waste and chemical inventories.
- Worked in conjunction with APHIS Legislative and Public Affairs to develop USDA-APHIS Program Aid No. 1996, "Attention Baled Hay Producers: Don't Transport Imported Fire Ants" (issued in February 2009).
- Chinese delegates from the General Administration of Quality Supervision, Inspection, and Quarantine of the People's Republic of China visited the United States to learn more about imported fire ants. PPQ's Emergency and Domestic Programs (EDP) and Phytosanitary Issues Management Team were the lead hosts for the visit, and CPHST scientists accompanied the group on visits to the CPHST Gulfport Lab, the port of Mobile, and the USDA Agricultural Research Service (ARS) Imported Fire Ant Lab in Gainesville, FL.
- Worked with a chemical company to prepare label language changes for a new insecticide dip treatment (bifenthrin) to be used for balled-and-burlapped nursery stock in the IFA quarantine. In fall 2008, the U.S. Environmental Protection Agency (EPA) approved the label, and EDP initiated changes to the Federal IFA quarantine to add a new treatment option for growers. During the last 5 years, testing to support this treatment option was conducted at the Gulfport Lab.

CPHST Lab, Mission, TX

Contact—Dr. Paul Parker

How We Support the Mission

As a result of several new pest and disease threats, the CPHST Mission Lab's focus has shifted over the past few years from one solely dedicated to biological control to a laboratory that is multifaceted in developing new methods in

molecular biology, integrated pest management, plant disease epidemiology, and biological control in support of PPQ programs. Because of these changes, the lab continues to grow in these new directions. For example, the Mission Lab's studies support fruit fly sterile insect technique (SIT) diet work and fruit fly trapping and monitoring. These efforts are geared toward providing more effective SIT management options. In 2008, we continued to evaluate diagnostic methods for important fruit fly pests, including *Anastrepha*, *Bactrocera*, and *Ceratitis* species. Offshore initiatives nearing completion include integrated pest management studies of chilli thrips (*Scirtothrips dorsalis*), giant African snails (*Achatina fulica*), and Cuban slugs (*Veronicella cubensis*). The offshore work dovetails nicely with ongoing molecular diagnostic work occurring at the lab with chilli thrips and gastropods.

In addition, monitoring continues to track populations of the Asian cycad scale, *Aulacaspis yasumatsui*, on sago palms in the Rio Grande Valley. For citrus, the current plant health issue focus is on HLB. As such, the Mission Lab is collaborating with the Texas A&M Citrus Center in Weslaco, TX, on a pilot, areawide management program for HLB's vector, the Asian citrus psyllid (*Diaphorina citri*) (fig. 15). In Florida, citrus canker work continues in collaboration with ARS personnel with special emphasis on infected fruit.

Recent Accomplishments

- A novel LBAM diagnostics enhancement included the development of a tortricid DNA barcode tool using museum or pinned specimens, which included generation of a database of over 300 DNA sequences of LBAM from around the world to enhance the LBAM DNA barcoding and compare genetic variation over its range.
- Identified and developed an additional DNA marker for LBAM diagnostics based on a PCR-agarose gel electrophoresis (AGE) method.
- Processed 14 geographic populations of the Mexican fruit fly using the inter-simple sequence repeat-PCR technique, which allows the genetic variation in the microsatellite DNA to be studied.
- Recovered the biological control agent *Gratiana bolivi-ana* on tropical soda apple in east Texas. An additional 450 natural enemies were released in May 2008.
- Completed a giant African snail molluscicide trial, including a bio-assay and field trial in Barbados.

- Completed a trapping test examining the quantity of Mexican fruit flies captured when different substances were utilized as an attractant. The substances included torula yeast and propylene glycol solutions, torula yeast and water, and two component biolures with propylene glycol solution. Torula yeast with Prestone Low-Tox propylene glycol captured significantly fewer flies than the torula yeast with water.



Figure 15. Asian citrus psyllid, *Diaphorina citri* (CPHST photo, Lisa Kennaway, Fort Collins, CO).

CPHST Lab, Otis Air National Guard Base, Cape Cod, MA

Contact—Vic Mastro

How We Support the Mission

The Otis Lab's mission is to identify, develop, and transfer technology for survey, exclusion, control, and risk assessment for APHIS and its cooperators. The lab serves a wide variety of PPQ programs that include: exotic pest-detection programs, such as CAPS; agricultural quarantine inspection (AQI) commodity treatments; and, emergency response and eradication programs for ALB (fig. 16), LBAM, Asian gypsy moth (AGM), emerald ash borer (EAB) (fig. 17), *Sirex noctilio* woodwasp, and other pests. Otis personnel identify high-risk exotic pests and develop survey technology to facilitate early detection of introductions. The lab continues to support the AGM program by creating risk maps, developing improved estimates of mate finding at low population densities, and producing the gypsy moth virus product, Gypchek.

Additional work is focused on the development of regulatory treatments for various commodities and means of their conveyance, such as pallets and containers. The Otis Lab is developing rearing systems for ALB and the winter moth (*Operophtera brumata*) and has biological control programs for the winter moth, EAB, and *S. noctilio*.



Figure 16. Asian longhorned beetle (Photo by Dave Lance, CPHST, Otis, MA).



Figure 17. Emerald ash borer (Photo by Dave Lance, CPHST, Otis, MA).

To fulfill its mission, the Otis Lab personnel maintain cooperative relationships with ARS, USDA's Forest Service, universities, and private industry. These cooperative arrangements extend to government organizations and universities in a number of foreign locations, including Australia, China, Japan, Korea, New Zealand, Russia, and South Africa. The work includes developing methods to monitor and exclude AGM from North America, predicting invasiveness of organisms by assessing damage on expatriate North American plants in foreign locations, developing attractants, and developing control techniques for targeted exotic pests.

Recent Accomplishments

- Supported the LBAM program by leading a technical working group and developing and evaluating control treatments. In particular, we participated in the planning of, and oversaw for PPQ, a pilot-scale field trial in New Zealand to evaluate several aerially applied mating disruption formulations for LBAM. These formulations release the female moth's sex attractant pheromone, which reduces LBAM populations by making it difficult for male moths to locate and mate with females. During the testing, we also monitored release of the pheromone over time under field and laboratory conditions. These tests identified two formulations that were effective enough to be recommended for use by the LBAM program.
- Collaborated with Australian scientists to develop and test new insecticide treatments for LBAM on nursery stock. These treatments allow nurseries in LBAM-infested areas of California to sell plants and ship them to uninfested areas. In comparison to previous treatment options, the new treatments are less toxic to humans and safer for the environment.
- Released exotic EAB parasites in several plots in Michigan in 2007 and 2008 and recovered parasites in the spring of 2008, indicating that they had successfully reproduced in the field. Otis Lab personnel helped with plans to develop a parasite production facility and release and evaluation programs, and provided hands-on instruction on rearing. The Otis Lab also developed a trap-lure combination that was used nationally by the EAB program in 2008 for its survey work. Our field projects on EAB management included evaluations of EAB dispersal and the systemic insecticide emamectin benzoate in West Virginia. We also organized an EAB Science Panel meeting in December 2008.
- Continued to perform DNA analysis of gypsy moth specimens submitted from PPQ's domestic monitoring program. The basic analytical procedures are now routine, and we are working to develop a protocol suitable for turning this function over to PPQ National Identification Services. This will allow reallocation of resources to continue work to develop new markers and a universal library of *Lymantria* specimens to provide better tools for identifying and determining geographic sources of exotic *Lymantria* specimens that are intercepted or detected in the United States.
- Continued assessing ALB populations and tree-damage patterns to understand the dynamics and spread of ALB infestations in New York, New Jersey, and Massachusetts. We consulted with the ALB program regarding the age, spread, and survey methodology for the recently discovered infestation in Worcester, MA.
- Formulated and provided over 125,000 pheromone dispensers to support the CAPS program and other survey efforts, including LBAM (outside of California) and false codling moth (*Thaumatotibia leucotreta*). We continued to provide advice on trap, lure, and survey design for these programs, and have arranged for a collaborator to synthesize the pheromone lure for rosy gypsy moth (*Lymantria mathura*), which is not commercially available.
- Conducted experimental releases of a biocontrol agent for *Sirex* woodwasp using the nematode *Beddingia siricidicola* in New York and Michigan. Studies to develop the methodology and timing for establishing trap trees for nematode release are already yielding results. Studies are also underway to characterize possible effects of the nematode on native woodwasps and their natural enemies.
- Development of regulatory treatments for pine wood, ash, and firewood is progressing. Good progress was made toward validating existing chipping protocols and International Standards for Phytosanitary Measures (ISPM-15) for heat-treatment schedules for *Sirex* in roundwood. Radio-frequency waves are also showing promise as a means of heat-treating wood.

CPHST Lab, Phoenix, AZ

Contact—Dr. Richard Zink

How We Support the Mission

The CPHST Phoenix Lab's mission is to develop, adapt, and implement areawide control technologies for new and existing program pests. Current work includes developing control methods for LBAM and pink bollworm (PBW), eradication programs using biotechnology, sterile insects, pheromone delivery systems, application technology, and geographic information system (GIS) applications. Scientific staff conduct extensive field studies to test materials and validate methods.

The Phoenix Lab's rangeland section interacts with Federal cooperators, and clients such as State and regional PPQ officers to foresee and identify problems or areas of needed improvement and to aid in our cooperators' program implementation. The rangeland section also envisions, develops, and implements solutions to problems and continuously evaluates all projects the program uses for optimization and use of current technology and tools for maintaining state-of-the-art status.

The lab uses specialized equipment, including: aerial application technology and equipment, environmental chambers and portable mass-rearing modules, a twin-screw extruder for insect diet development, a room that allows for the simulation of aerial applications of sprayed products, a quarantine laboratory for rearing genetically modified PBW, and equipment for testing pesticide and pheromone application technology.

In addition, Phoenix Lab personnel work closely with the PBW Rearing Facility to develop and test mass-rearing technology for implementation of improved methods on a large scale. They also work cooperatively with PPQ field personnel to evaluate operational-scale grasshopper and Mormon cricket treatments.

Recent Accomplishments

Pink Bollworm

- Modified a recently developed sprayable PBW mating-disruption formulation so that it can be applied aerially. This formulation can be extended to other invasive pest species that infest crops or rangeland.
- Acquired, cataloged, and made thousands of books, technical bulletins, circulars, university-published documents, pamphlets, Web sites, maps, and articles concerning PBW available to researchers. The unpublished writings and data for PBW researchers will also be added to the archive.
- Tested possible additives to mating disruptant to make the formulation thicker for aerial application without it directly interfering with its chemical attractiveness or longevity. We used an inexpensive method of placing various additives—mixed with the mating disruptant in PBW traps—in fields with measurable, sterile PBW populations. The work was completed in two agronomically different cotton-growing areas over the span of the season.

- Developed a new egg disinfectant that provides an environmentally friendly substitute for formalin in the PBW Rearing Facility. The new treatment will be used in the 2009 sterile insect production season for the eradication program.
- In collaboration with CPHST Gulfport Lab, Gulfport, MS, we evaluated and documented calco red dye concentrations in the rearing facility artificial diet. We utilized a CARY UV/Visible Spectrophotometer™ to determine calco red dye levels. We also evaluated calco red dye presence in PBW moths reared on artificial diet.
- Conducted the first open-field test of a PBW sterile marker strain genetically modified to express a fluorescent protein for potential use in the eradication program. Use of a genetic marker strain of PBW would allow error-free identification of released moths in monitoring traps. That, in turn, will improve program effectiveness.
- Designed and built a specialized stand-alone facility for mass-rearing and collecting quarantine-related moths. This facility can produce more than 500,000 genetically modified sterile PBWs a week for field testing. In addition, this rearing unit could serve as a prototype for mass production of other lepidopteron pests.

Grasshopper and Mormon Cricket

- Further developed an economical system to evaluate potential fungal pathogens for rangeland grasshoppers. The system consists of technology that simulates aerial field application by spraying from a tower and field cages designed to allow normal thermoregulation of grasshoppers.
- Evaluated new solid baits as candidate alternatives for use in Mormon cricket and rangeland grasshopper control programs. Additional useable baits ensure the availability of the solid bait option for both ground aerial applications and are particularly important in continuing the hot spot treatment philosophy.
- Began reducing, simplifying, and standardizing oil and water diluents used in diflubenzuron treatments. These changes will lead to reduced drift, greater deposition, and simpler, cheaper treatments, and they may also cause further reductions in active ingredient use with this formulation.
- We were instrumental in initiating a western, 17-State, comprehensive soil survey to detect and isolate potential insect fungal pathogens. The survey results will be used

in the eventual development of biological insecticides for Mormon cricket and rangeland grasshopper control. Results from these efforts will also have implications for current and future pests.

CPHST Plant Epidemiology and Risk Analysis Laboratory, Raleigh, NC

Contact—Robert Griffin

How We Support the Mission

The CPHST Plant Epidemiology and Risk Analysis Laboratory (PERAL) is PPQ's primary unit producing pest risk analyses (PRA). In this laboratory, a diverse group of scientists and professionals provides essential scientific support to risk-based policymaking across a broad range of phytosanitary issues. Staff members use sound science to analyze both import and export issues and facilitate safe trade. The PRA help safeguard American agriculture and plant health from harmful exotic plant pests in both managed and unmanaged ecosystems. More specifically, the analyses help PPQ to design risk-based regulations for import and domestic pest management programs, identify and assess new pest threats, monitor the effectiveness of existing programs, and optimize available resources to enhance protection. PERAL personnel also provide technical support documents PPQ requires for pests, commodities, and pathways. These products may include risk maps that indicate existing or potential range domestically or internationally or that predict ranges from weather- or climate-matching analyses.

Over the years, PERAL has established itself as a global leader in both productivity and quality management. PERAL is currently the only International Standards Organization (ISO)-certified plant health risk analysis unit in the world. The group contributes significantly to the promotion of international dialogue and increased capacity for science-based management of phytosanitary issues through its Risk Analysis Mentoring Program (RAMP) for visiting scientists. In addition, PERAL provides basic PRA training workshops, with topics covering the spectrum of concepts, methods, and resources associated with pest risk analysis. PERAL also provides training (regulatory curriculum) to students in relevant fields on key aspects of regulatory plant protection through cooperative relationships with academic institutions.

Furthermore, PERAL promotes regional and international harmonization of plant health regulations by participating in the North American Plant Protection Organization (NAPPO) and the International Plant Protection Convention (IPPC).

Recent Accomplishments

- Finalized 18 original and 30 revised PRA in 2008—facilitating safe trade for 46 commodities from 58 different countries, or a total of 119 new country-commodity combinations.
- The New Pest Advisory Group (NPAG) analyzed 33 pests and completed an additional 18 preassessments for organisms that were not considered NPAG pests.
- The Exotic Pest Information Collection and Analysis (EPICA) team produced 38 notifications containing articles on pests of regulatory significance.
- PERAL staff processed risk assessment and informational documents for the organism, pathway, export, and risk mapping areas, including: 2 organism, 3 pathway, 29 export, and 7 risk mapping documents.
- The 6-member Caribbean Pathway Analysis team drafted a 250-page document with 9 chapters.
- The Global Pest and Disease Database (GPDD) team added 208 new pests and updated an additional 384 pests in the database. GPDD was expanded to accommodate EPICA, incorporating 49 EPICA notifications and 260 articles. The team also incorporated 12 new forms and tables in preparation for adding the new PRA archive tool.
- The PERAL Weed Team developed a new weed risk assessment system similar to the Australian system and is currently validating the model for use in the United States.

Biological Control Unit, Raleigh, NC

Contact—Dr. Ken Bloem

How We Support the Mission

The Biological Control Unit (BCU)—a virtual team of 10 to 12 CPHST scientists—focuses on developing technologies that allow living biological organisms (e.g., natural enemies and competitors) to lessen the impacts of introduced, inva-

sive insect pests, weeds, and plant pathogens. Use of these organisms also minimizes impacts on the environment and non-target organisms. The BCU functions within CPHST's Survey Detection and Identification national program. The BCU strives to better prepare the agency for potential U.S. exotic pest incursions and provide effective pest-management options. BCU scientists provide technical oversight and expertise to programs to ensure that scientific knowledge gaps are identified and addressed and that developed technologies are quickly transferred to stakeholders. The BCU also provides support in the discovery and evaluation of new biological control agents occurring offshore and domestically as well as develops cost-effective rearing and monitoring systems for approved biological control agents and their hosts. In addition, the BCU provides permitted biological control agents collected from established field insectaries and laboratory colonies to PPQ project managers and other project cooperators, ensures the safety of biological control agents by continued post-release monitoring, and develops educational and programmatic materials for use by PPQ and other collaborators.

Recent Accomplishments

- Documented that *Spathius agrili*, a parasitoid of EAB, can reproduce and overwinter in Michigan following field releases and transferred technology for mass rearing *S. agrili* to the new regional rearing facility in Brighton, MI.
- Discovered new natural enemies of EAB—including additional species of *Spathius*—in South Korea, China, and Russia. Collections are being forwarded to the Otis Lab quarantine facility for rearing.
- Isolated a naturally occurring strain of the nematode biocontrol agent (*Beddingia siricidicola*) of *Sirex* in New York and had it identified by DNA analysis.
- Mass-reared nematode biocontrol agents of *Sirex* in the Otis Lab and conducted experimental field releases against *Sirex noctilio* in New York and Pennsylvania.
- Characterized an isolate of *Albugo candida*, which causes white rust, collected from perennial pepperweed (*Lepidium latifolium*). Results indicate that this pathogen may be a new race because it is not 1 of the 11 reported races occurring on a range of weeds and crop species in the United States. Currently, host specificity testing with this isolate and an isolate from California is underway.
- Planned and implemented a survey to determine the distribution of the tropical soda apple (TSA) biological control agent, *Gratiana boliviana*, in Florida as a cooperative effort with scientists and government personnel from ARS, the University of Florida, and the Florida Division of Plant Industry to decide if further mass production and releases are warranted.
- Conducted releases of the biological control agent *G. boliviana* on TSA in eastern Texas during the summers of 2007 and 2008 and documented that the beetle can overwinter and survive under Texas conditions.
- Developed a ribosomal DNA ITS2 marker that can be utilized to detect chilli thrips DNA from predatory insects. The marker has been used to detect the pest's DNA in several predatory species, including: *Orius insidiosus*, *Amblyseius swirskii*, and *Chrysoperla rufilabris*.
- Initiated a study of the coffee mealybug (*Planococcus lilacinus*)—an invasive pest of citrus, grape, potatoes, mangos, and guava—in the Dominican Republic to determine its potential impact and search for local natural enemies.
- In collaboration with cooperators at Florida A&M University, determined the efficacy of two passion-vine mealybug (*Planococcus minor*) parasitoid species, *Leptomastix dactylopii* and *Coccidoxenoides perminutus*, which were observed controlling the pest in Trinidad. Additional experiments were conducted to determine the temperature tolerance of the passionvine mealybug in order to predict its possible distribution if it becomes established in the United States.
- Evaluated the impact of local natural enemies of red palm mite in Trinidad and determined that—despite dramatic increases in predator numbers—the local natural enemies did not reduce populations of red palm mite. This finding indicates the need for the introduction of exotic natural enemies.
- In collaboration with researchers at the University of Florida's Tropical Research Education Center, determined that populations of the invasive scale *Crypticerya genistae*—a potential pest of soybeans, peanuts and other legumes—were reduced by more than 95 percent in 1 year on Miami study sites. A predatory beetle *Anovia circumclusa*, recorded for the first time in North America as prevalent at all sites, readily controlled the pest in laboratory studies and could serve as a biological control agent if the pest moves to other States.

- Developed laboratory rearing procedures for *Tamarixia radiata*, a parasitoid of the Asian citrus psyllid, and examined efforts needed for potential mass-rearing in support of field programs. Studies were initiated to determine parasitism rates and host-feeding effects in the laboratory and field.
- Monitored populations of Asian cycad scale on sago palms in Texas and discovered high levels of a parasitoid attacking the scale. The parasitoid was identified as belonging to the *Aphytis lingnanensis* group. This is the first report of an *Aphytis* species attacking this scale.
- Provided more than 70,000 adult saltcedar leaf beetles (*Diorhabda elongata*) collected in western Nevada for release at field insectaries in Colorado, Iowa, Kansas, Nebraska, Oregon, South Dakota, Washington, and Wyoming.
- The gall wasp *Aulacidea acroptilonica*, the first insect to be released as a biological control agent of the exotic weed Russian knapweed (*Acroptilon repens*), was permitted for U.S. release in 2008; initial field releases will be made in Wyoming in 2009.
- Established a laboratory colony of LBAM and provided host material (eggs, larvae, pupae) to University of California and California Department of Food and Agriculture cooperators investigating LBAM biological control.
- Located in central Turkey a potential new biocontrol agent for *Tamarix* (salt cedar), *Liocleonus clatratus*, and collected 53 adults for initial studies that took place in Rome in cooperation with the Biotechnology and Biological Control Agency.

Treatment Quality Assurance Unit, Raleigh, NC

Contact—Scott Wood

How We Support the Mission

The Treatment Quality Assurance Unit (TQAU) safeguards American agriculture and natural resources by developing, adapting, and supporting technology to detect, identify, and mitigate the risk posed by exotic pests in preclearance programs and at ports-of-entry. APHIS' last line of defense in preventing an exotic pest invasion is the commodity treatment that mitigates this risk. Currently, core activities

within TQAU include developing quarantine treatments, contributing to the treatment manual for ports-of-entry, certifying shipping containers and vessels, maintaining the Commodity Treatment Information System (CTIS), training-the-trainer for proper application of treatments, certifying commodity treatment facilities within foreign countries, and conducting quality assurance audits of treatments performed domestically and at foreign sites.

Recent Accomplishments

- Implemented a new Niger seed database component within the CTIS. This is a continuing endeavor to capture data from all phytosanitary treatments performed both nationally and internationally.
- Continued support and expansion of the PPQ Form 429 fumigation database, PPQ Form 556 cold treatment database, and the Irradiation Reporting and Accountability Database for APHIS and industry use. These systems are critical to supporting phytosanitary treatments around the world.
- Reviewed new commodity treatment research information for high-risk pests, including the Asian fruit fly (*Bactrocera invadens*), Queensland fruit fly (*Bactrocera tryoni*), Chilean false red mite (*Brevipalpus chilensis*), Mediterranean fruit fly (*Ceratitidis capitata*), and LBAM (*Epiphyas postvittana*).
- Performed audits of nonchemical and chemical treatments (e.g., methyl bromide treatments). Failure analysis was performed on cold treatments shipped in containers.
- Certified and approved irradiation facilities in Thailand, Vietnam, and Mexico. The following fruits were approved for irradiation: mangosteen, longans, mangoes, rambutan, and guavas from Mexico, pineapple from Thailand, and dragon fruit from Vietnam.
- Implemented new thermal mapping techniques for vapor heat and forced hot air treatments in Mexico and Hawaii. Established an Excel-based data capture system for processing and analyzing data from thermal mapping.
- Completed electronic indexing of 6,000 documents in the TQAU library in an effort to develop a paperless electronic system for AQI treatments, policies, and research literature. This multiple-year process has accumulated a grand total of 14,000 documents.



Project Highlights

Development of Proficiency Test Reagents for High-Consequence Plant Pathogens for Delivery to NPPLAP for Laboratory Certification

Location: CPHST Lab, Beltsville, MD

Lead Scientist: Dr. Vessela Mavrodieva

Team Member: Sarika Negi

The *Phytophthora ramorum* laboratory provisional approval program led by the Center for Plant Health Science and Technology (CPHST) began in 2005 in response to a Federal Order requiring certification of plant material from California, Oregon, and Washington. The Federal Order also required molecular testing of thousands of samples for *P. ramorum*, which would overwhelm CPHST laboratory capacity due to the volume of samples. Increased capacity for regulatory plant pathogen diagnostics was needed, so APHIS-Plant Protection and Quarantine (PPQ) engaged the newly developed National Plant Diagnostic Network (NPDN) and State department of agriculture laboratories using lab accreditation and diagnostician certification to provide needed diagnostic capacity. The initial *P. ramorum* proficiency testing (PT) panels were developed and distributed for nine laboratories in 2005. Since then, over 30 laboratories have participated as part of the National Plant Protection Lab Accreditation Program (NPPLAP) effort to ensure high quality and scientifically defensive diagnostic results for regulatory plant pathogens required by the PPQ domestic and emergency programs. The PT program has matured in 2008 with the implementation of a citrus greening (also known as huanglongbing [HLB]) PT panel to certify labs in citrus-growing States.

Proficiency testing is an integral part of laboratory accreditation programs to: (1) provide external and independent assessment of the accuracy of the results generated by each participant and (2) verify that the performance of each test site is comparable to other labs performing the same analysis. Successful PT analyses mimic all steps of the diagnostic process, including both tissue-based and DNA-based sample preparation. To develop and rigorously validate panels, we included plant material for extraction and testing by polymerase chain reaction (PCR). To overcome major challenges, we developed and validated a PT panel from which the pathogen could be detected at different levels from plant material that maintained long-term performance characteristics (fig. 18) and a determination of suitable storage conditions for the preserved tissue.



Figure 18. *P. ramorum* PT08 tissue panel samples. Lyophilized infected and/or healthy rhododendron tissue in vacuum sealed vials destined for labs participating in the PT program (Photo by Vessela Mavrodieva, CPHST, Beltsville, MD).

We cooperated extensively with Dr. Paul Tooley, Research Plant Pathologist with the USDA-Agricultural Research Service's Foreign Disease-Weed Science Research Center in Ft. Detrick, MD, in a project to develop and provide infected plant materials for *P. ramorum* testing that are suitable for PT panel development. Once developed, the tissue-based panels were monitored monthly and showed excellent stability (fig. 19). The distribution of well-developed and validated stable

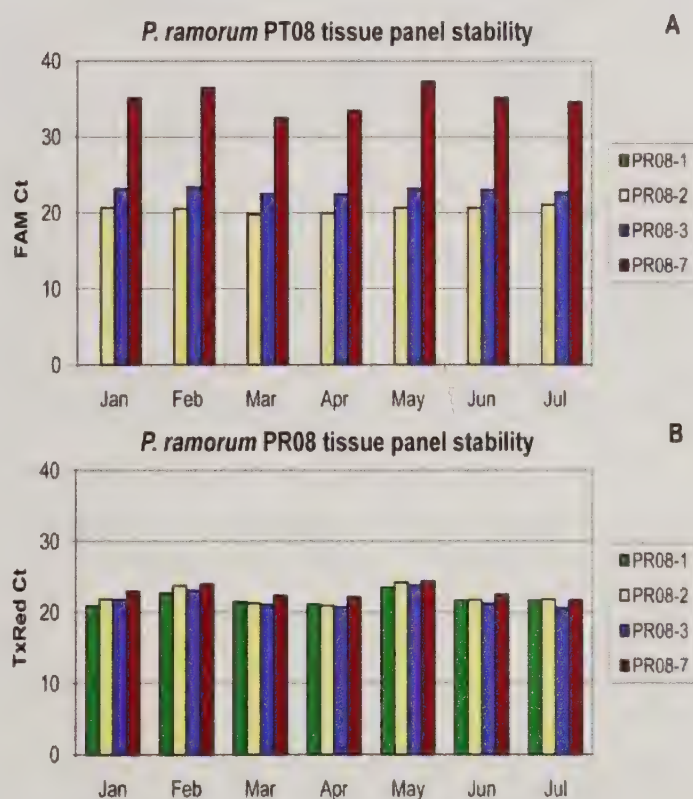


Figure 19. Demonstration of the *P. ramorum* tissue panel stability. The stability of the tissue panel was monitored monthly using real-time PCR following DNA extraction. Panel A demonstrates the ability to detect the pathogen, and the quality of DNA extraction is demonstrated by detection of an internal control in panel B (Image by Vessela Mavrodieva, CPHST, Beltsville, MD).

panels—along with clearly written sample preparation and PCR standard operating procedures—contributed to the demonstration of high proficiency by participating analysts certified in fiscal year 2008 (fig. 20).

Accreditation of labs and certification of diagnosticians by PT testing helps to ensure preparedness and response. Additional laboratories were certified for *P. ramorum* testing in 2008. The CPHST Lab and NPPLAP developed tissue-based panels, approved eight laboratories for HLB diagnostics, and contributed to preparing all citrus States for response to the threat of HLB.

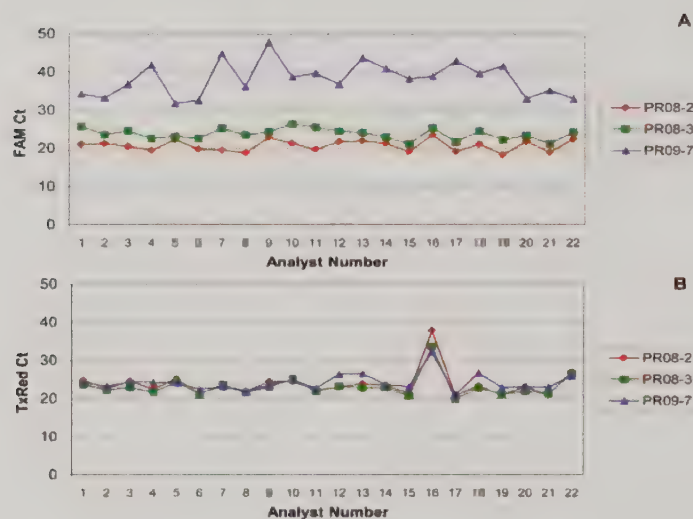


Figure 20. *P. ramorum* PT08 tissue panel results tested in real-time PCR by 21 participants for 3 tissue samples (PR08-2, PR08-3, and PR08-7). Ability to detect the pathogen is demonstrated in panel A, and quality of DNA extraction is demonstrated by detection of an internal control in panel B (Image by Vessela Mavrodieva, CPHST, Beltsville, MD).

Robotic Automated Pest Identification

Location: CPHST Lab, Fort Collins, CO

Lead Scientist: Dr. Jeff Drake

The Robotic Automated Pest Identification (RAPID) project strives to deliver a multifaceted survey support system. By automating the handling, classification, physical sorting, and reporting of survey samples, RAPID aims to greatly reduce the volume of survey samples that must be processed and identified by hand (fig. 21). Beyond the advantages of speed and relief from tedium, automation captures data instantly and frees limited and highly trained human resources to

focus on unusual finds as well as the broader survey objectives. RAPID utilizes the latest advances in image processing and analysis, remote sensing, pattern recognition, and industrial robotics technology to provide fast, automated identification in support of targeted pest species detection.

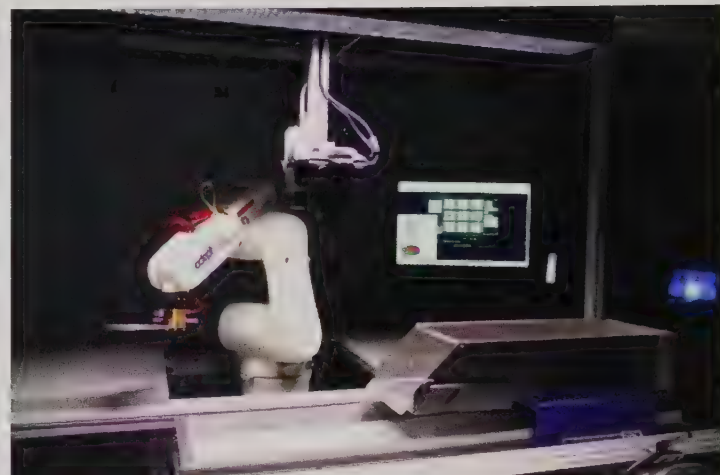


Figure 21. RAPID (Robotic Automated Pest ID) technology includes robotics, automated sample feeding, image analysis, and relational databases. RAPID ALPHA, shown here, is utilized in the development and testing of all survey support tools in the project (Photo by Jeff Drake, CPHST, Fort Collins, CO).

Many pest survey and detection processes, where the same tasks are continually repeated, lend themselves well to the application of industrial automation techniques. These are the same basic techniques of automated material handling and image analysis that were so successfully applied to automated genome surveys. RAPID tools are being developed to support a wide variety of survey requirements, including pest detection, predator and prey quantification for integrated pest management (IPM), and commodity seed purity analyses. RAPID is a Federal/State collaborative effort among CPHST, USDA's Forest Service, and New Mexico State University's (NMSU) College of Agriculture.

During 2008, two tools—Pest Identification Spectral Camera Experiment Station (PISCES) and Robotic Information Technology Assistant Web Interface (RitaWeb)—were designed, developed, and tested for the RAPID System. From the project's inception, we recognized that attempting to automate traditional taxonomic identification, such as appraising sutures and hairs, was not generally a practical approach for automation. Two fundamental objectives of the RAPID project have been to develop new, nontraditional diagnostic techniques and to identify traditional techniques suited to

automated survey. For example, this past year, the RAPID team advanced the development of a diagnostic capability to evaluate the reflected optical radiation from samples; the capability is now called PISCES.

PISCES is a hyperspectral imaging spectrometer that is unique in its capability to optically image a group of samples, providing their morphological and spectral properties (400–1100 nanometers [nm]) for diagnostics (fig. 22). Another important spectrometry tool that is part of PISCES is an ASD Inc. fiber optic point spectrometer, which extends the spectrum that can be evaluated much further into the near infrared (NIR), up to 2500 nm. Absorption bands can be expected in this NIR. These absorption bands arise from the fact that compounds vibrate with often unique frequencies in the NIR range, forming a sort of spectral fingerprint. These spectral fingerprints, not visible to the human eye, hold significant potential as diagnostics. In 2008, the PISCES spectrometry tools were brought on board to test whether the potato cyst nematode's (PCN) spectral signature was distinct from other morphologically similar entities resulting from processed soil samples.

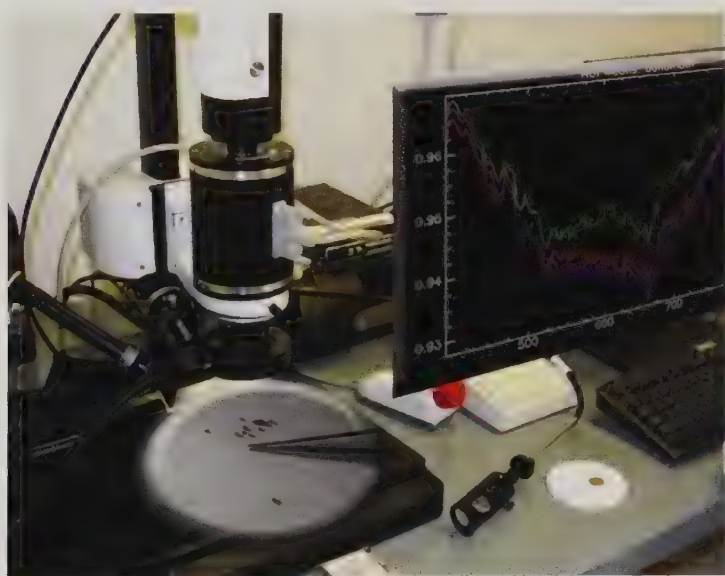


Figure 22. Project scientists use PISCES (Pest Identification Spectral Camera Experiment Station) to investigate multi-spectral properties of three different nematode cysts for diagnostic features (Photo by Jeff Drake, CPHST, Fort Collins, CO).

RitaWeb forms the data foundation for the suite of RAPID survey support tools. For the first time, users of the RAPID automated survey support tools have access to a complete electronic, end-to-end survey support system (fig. 23). RitaWeb is composed of Web-based, server-side applications and relational databases that link all aspects of survey, providing instant access to survey data. RitaWeb links all RAPID survey tools and steps—from the definition of survey parameters, such as trap location and date of collection, through automated identification and documentation via imaging, to the generation of survey statistics and reports. RitaWeb leverages the power of the industrial automation techniques that are fundamental to RAPID, automatically documenting data on every sample that enters the system. RitaWeb also provides immediate, easily accessible, and powerful new information for managers, decisionmakers, survey personnel, and scientists.

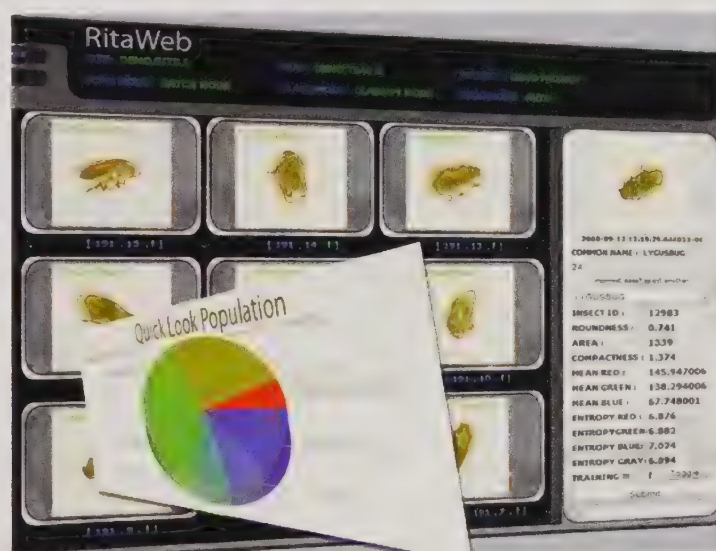


Figure 23. RitaWeb (Robotic Information Technology Assistant Web Interface) provides timely, Web-based access to powerful new information for managers, decisionmakers, survey personnel, and scientists (Image by Jeff Drake, CPHST, Fort Collins, CO).

In 2009, the RAPID team intends to deliver a successful pilot study to the USDA Forest Service and cooperators that will demonstrate the RAPID system's ability to support wood boring beetle surveys. Additionally, the team will continue its efforts to identify automation techniques in support of PCN surveys and will provide support to NMSU's survey efforts for IPM in agricultural crops.

Rapid Development of Matrix-Specific Imidacloprid Detection Methods Supporting the Emerging Asian Longhorned Beetle (ALB) Program in Massachusetts

Location: CPHST Lab, Gulfport, MS

Lead Scientists: Lisa Mosser and Robert D. Smith

In the fall of 2008, the APHIS Environmental Compliance Team requested this emergency project to assist in developing a new ALB chemical treatment program in Massachusetts. A recently detected, serious outbreak of ALB in the area prompted the consideration of chemical treatment suppression options using imidacloprid. The proposed treatments were to begin in the spring of 2009. Local beekeepers and honey and sap producers needed immediate assurance that their crops and commodities would not be impacted by a Government-applied chemical treatment program. Analytical methods were rapidly developed for the detection of imidacloprid in honey, tree sap, bees, and bee's wax matrices (fig. 24). Although every matrix requires a different process, each method utilizes a complex liquid/liquid extraction and concentration preparation (fig. 25), coupled with high-pressure liquid chromatography analysis and confirmation.

Eighty field samples from the proposed treatment area were submitted as controls and analyzed to establish a baseline prior to the beginning of scheduled treatments in the spring of 2009. All methods-development work, related sampling, and sample analysis was conducted in less than 45 working days. As a result of this project, APHIS is now prepared to conduct routine sampling and analysis to ensure that the ALB chemical treatments applied do not adversely impact the local bee and syrup industries. In partnership, the CPHST Gulfport Lab's Analytical Chemistry section and the APHIS Environmental Compliance Team can now provide clear and relevant monitoring data to all local bee and syrup industry leaders, which helps ease concerns and return the focus to the control and eradication of the ALB pest in the area.



Figure 24. New ALB program specific matrices: honey, bees, bee's wax, and tree sap (Photo by Lisa Mosser and Robert D. Smith, CPHST, Gulfport, MS).



Figure 25. Liquid/liquid primary extraction matrix specific for honey (Photo by Lisa Mosser and Robert D. Smith, CPHST, Gulfport, MS).

National Panicle Rice Mite Survey 2008

Location: CPHST Lab, Mission, TX

Lead Scientist: Josie Salinas

The panicle rice mite (PRM), *Steneotarsonemus spinki*, is considered the most destructive mite pest affecting rice around the world. The mites are clear to straw-colored and approximately 250 micrometers (μm) in length, making them very difficult to identify with the untrained eye (fig. 26). They pose a threat to rice crops in the United States, where yield losses can range from 30 to 90 percent. Because the PRM feeds on the rice plant behind the leaf sheath (fig. 27) and inside the seed's endosperm, it can be very difficult to recognize its presence and manage with chemical control practices.

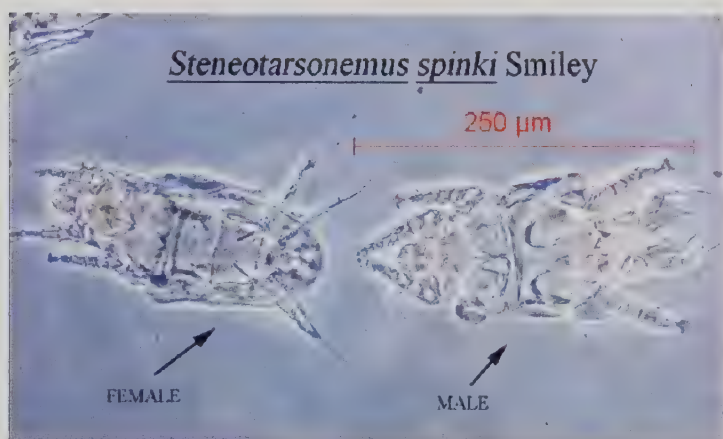


Figure 26. Adult panicle rice mites under compound microscope (Image by Eric McDonald, PPQ, Humble, TX).

As outlined in the New Pest Response Guidelines developed after the initial U.S. detection of PRM, a national survey conducted in rice-growing States was implemented. The CPHST Mission Lab was asked to provide technical support in the survey. Samples were received and processed at the Arthropod Quarantine Facility, which is part of the Mission Lab.

In early 2008, Mr. Eric McDonald, a PPQ authorized mite identifier, trained CPHST personnel on how to perform plant material washings for extractions, screen suspect mites from rice samples, and identify PRM. The standard operating procedure is highly technical and requires meticulous



Figure 27. Panicle rice mite eggs and adults feeding inside rice sheath. Male (center) surrounded by two females. Image magnified 1,000 times under stereo-microscope (Image by Eric McDonald, PPQ, Humble, TX).

techniques, such as sanitizing between samples, to prevent cross-contamination. Two technicians were assigned to the washing procedure and four individuals were assigned to the screening and identification of PRM and other morphologically similar mites. The survey team spent over 5,000 hours enforcing standard operating procedures.

All suspect mites were isolated on slides and forwarded to Eric McDonald in Houston for confirmation. First detections of PRM within a State were sent for final confirmation to Ronald Ochoa, the mite specialist at the Systemic Entomology Laboratory in Beltsville, MD.

In early December 2008, the survey—which was comprised of nearly 4,000 plant samples—was completed. Samples were received between March and November 2008 from 11 States, including Arkansas, Connecticut, Florida, Indiana, Louisiana, Maryland, Minnesota, Mississippi, Missouri, Ohio, and Texas. Samples were submitted both from fields and research greenhouses. All samples submitted from fields were negative for the presence of PRM. However, PRM was detected in samples collected from research greenhouses in Arkansas, Louisiana, and Texas. These research greenhouses are currently utilizing eradication techniques.

The PRM team (fig. 28) received recognition from the PPQ Eastern and Western Regions in November 2008 for providing critical technical support. The team was comprised of one CPHST scientist, two CPHST technicians, and four technicians funded through the Western Region.



Figure 28. PRM survey team: front row, Joana Cordoba and Albino Chavarria; middle row, Alizma Reyes, Daniel Martinez, and Michael Pena; back row, Josie Salinas and Antonio Perez, Jr. (Photo by Josie Salinas, CPHST, Mission, TX).

Implementing and Evaluating Biological Control of *Sirex* Woodwasp

Location: CPHST Lab, Otis Air National Guard Base, Cape Cod, MA

Lead Scientist: Dr. David W. Williams

Sirex woodwasp (*Sirex noctilio*) was first detected in Oswego County, NY, in 2004. The woodwasp is a destructive outbreak pest that kills plantation pines throughout the Southern Hemisphere. This exotic invader is not under effective natural control, is already widely distributed in eastern North America, and poses a serious threat to our pine forests if not controlled. It also moves rapidly; since the initial find, *Sirex* woodwasp has been discovered in Pennsylvania, Michigan, and Vermont. The potential for damage to our forest resources will be especially high when the woodwasp reaches the pine plantations of the southeastern and western United States.

Our approach to managing *Sirex* woodwasp is biocontrol. Its most effective natural enemy is a nematode (*Beddingia siri-cidicola*) that sterilizes woodwasp females. The nematode has an intriguing life history, with the ability to develop into two different physical forms. One form grows on the symbiotic

fungus carried into a tree when *Sirex* woodwasp lays eggs, and the other parasitizes woodwasp larvae. The nematode has been used successfully in management programs in the Southern Hemisphere. During the past 30 years, Australian scientists have developed technology for mass-rearing and delivering the nematode on a large scale. Because of the success of the Australian biocontrol program, we chose to adapt it for use in the United States. The Australian program schedules activities through three seasons. In the spring, trap trees are created by using herbicides to stress pines and attract woodwasp females to lay eggs. During the summer, nematodes are mass-reared commercially. Lastly, in the fall, trap trees are felled and inoculated with nematodes, which will infect many woodwasp larvae that then develop into sterile adults.

We have made considerable progress in transferring the biocontrol technology to the United States in the short time since woodwasp detection. We set up a lab for rearing nematodes in 2006. We received nematode cultures from Australia in 2006, have maintained them in the lab since then, and mass-reared nematodes (fig. 29) for field releases in 2007 and 2008. We carried out three nematode releases from 2006 to 2008. The goals of the releases were to test the Australian inoculation method, to assess the establishment of nematodes in American pines, and to evaluate overwintering survival of the nematodes in North America. The releases were “controlled”; that is, trees were inoculated in the fall (fig. 30), samples were collected in winter, and remaining tree materials were destroyed before woodwasps emerged. Controlled releases were necessary because of lingering concerns expressed by the environmental conservation community about the potential effects of these biocontrol efforts on native woodwasps. With knowledge gained from these experiments, we are now prepared to request permits for operational releases. Our deliverables include the ability to mass-rear nematodes in the laboratory, provide them when needed for wide scale releases, and apply them in the field.

We are grateful to our PPQ colleagues for their assistance. We acknowledge productive partnerships with several State agencies: the New York Department of Agriculture, New York Department of Marketing and Environmental Conservation, Pennsylvania Department of Conservation and Natural Resources, and Michigan Department of Agriculture. We thank our cooperators at Cornell University for

investigating basic questions arising from the project. Finally, we thank the Canadian Forest Service for its generous help in the molecular identification of nematodes and Mr. Robin Bedding, a retired Chief Scientist from Australia's Commonwealth Scientific and Industrial Research Organization, for his unstinting advice on all phases of the project.



Figure 29. Lab cart with 500 ml nematode rearing flasks. Each flask contains about 1 million nematodes (Photo by David Williams, CPHST, Otis, MA).

Evaluation of Aerial Application of a Long-Lasting, Sprayable Formulation of Mating Disruptant for Pink Bollworm Moth in Non-Bt Cotton

Location: CPHST Lab, Phoenix, AZ

Lead Scientists: Dr. Michelle Walters, Barry Barnes, Dr. Nelson Foster, and Nathan Moses-Gonzales

The pink bollworm (PBW) moth, *Pectinophora gossypiella*, has been a key pest of cotton across the southwestern United States and northern Mexico. It is currently the target of a



Figure 30. Punching holes for nematode inoculations (Photo by David Williams, CPHST, Otis, MA).

multinational, areawide eradication program that relies on mating disruption as one of its tactics. The standard mating disruption product, PB-Rope L, is effective and long-lived, but requires labor-intensive hand application. Since labor is in short supply, a mechanically applied mating disruption formulation that lasts at least 30 days is needed.

From 2003 to 2008, the eradication program utilized various methods, including aerial and ground applications, to apply pheromone to protect the cotton crop and reduce PBW reproduction. During this time, CPHST tested numerous formulations to ensure that the best options were available to the program. Formulations were tested under several field conditions, multiple locations, and with various application methods. In 2008, we used six nongenetically modified cotton fields. Three were treated with PBW-GEL that was supplied by Pacific Biocontrol, which is the most promising formulation to date, and three were left untreated. These fields received 2,700 sterile PBW moths per week per acre as part of the eradication program. We also placed traps containing lures and checked them twice a week in order to accurately gauge the PBW population in the fields. Large,

regularly spaced droplets (fig. 31) were achieved in previous research using a ground rig. Because of the air speed and wind shear during aerial application, we added 1 percent NALCO-TROL—a drift control additive supplied by Nalco Co. containing 30 percent polyvinyl polymer—to greatly thicken the GEL, so that the formulation would hold together in the air.

While our first application was hindered by various elements—namely 1 inch of pouring rain within 1½ hours of application—it was still effective for 21 days. Our second application proved highly effective; it lasted over 46 days, which exceeded our objective of discovering a formulation that lasted for at least 30 days (fig. 32). This improved formulation of PBW-GEL will aid the eradication program by reducing labor costs associated with applying pheromone and will support the farmer by increasing crop yield (as PBW populations will be disrupted over longer periods of time).



Figure 31. Pheromone droplets on cotton 2 weeks post treatment (Photo by Michelle Walters, CPHST, Phoenix, AZ).

Our thanks to our partners and collaborators external to CPHST, including: Jack Jenkins, Pacific BioControl, Litchfield, AZ; Timothy Roland and Daryl Hill, USDA-APHIS-

PPQ, Edinburg, TX; Larry Antilla, Arizona Cotton Research and Protection Council, Phoenix, AZ; and Robert Staten, USDA retired, Gilbert, AZ.

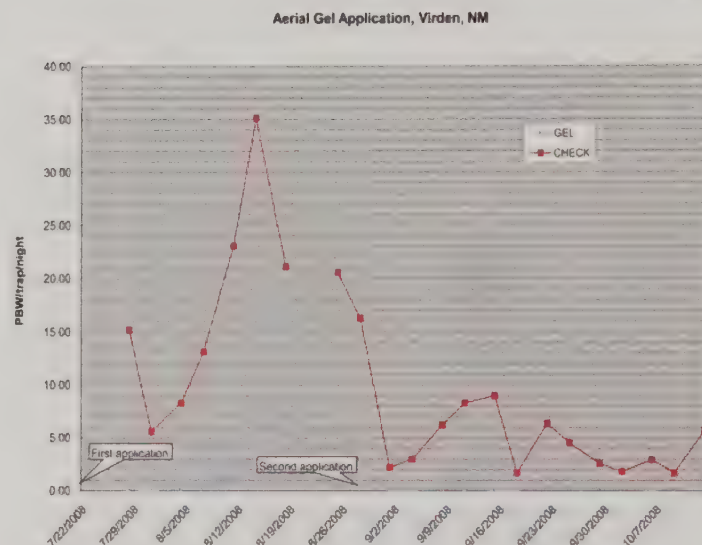


Figure 32. Results of two applications of a sprayable pheromone for pink bollworm (PBW GEL) versus untreated check, in Virden, NM, July through October 2008. Applications of PBW GEL maintained trap counts below the economic treatment threshold, one moth/trap/night, for 20 days following the first application and for 46 days following the second application. Differences from the check fields were significant for all days (even when the threshold was exceeded) (Image by Michelle Walters, CPHST, Phoenix, AZ).

Evaluation of Pathways of Exotic Plant Pest Movement Into and Within the Greater Caribbean Region

Location: CPHST Lab, Plant Epidemiology and Risk Analysis Lab, Raleigh, NC
Lead Scientist: Dr. Heike Meissner

The Greater Caribbean Region (GCR), including the U.S. Gulf States, increasingly suffers economic and environmental impacts due to the introduction of exotic plant pests. While we can only estimate how many exotic species have already established in this region, it is widely acknowledged that their number is in the hundreds and rapidly growing. The United States will continue to be at risk because (1) the land areas in the GCR share a similar climate and flora, and (2) exotic pests that become established in one part of the region are potentially able to invade most other parts.

The United States has made efforts to help coordinate the implementation of an effective regional safeguarding system,

contributing significantly to the Caribbean Regional Invasive Species Intervention Strategy (CRISIS) of the Caribbean Invasive Species Working Group (CISWG). However, a lack of understanding of pest movement's potential pathways has hindered prioritization and resource allocation decisions. To address this knowledge gap, the PPQ Executive Team funded a year-long project, which was carried out in collaboration with CISWG and implemented by PPQ risk analysts. The objective of this project is to use Government data, published scientific literature, and information collected during site visits to assess the likelihood of pest movement via various pathways into and within the entire GCR.

CPHST risk analysts coordinated with the APHIS Policy and Program Development (PPD) Risk Analysis Systems (RAS) office to implement the project. External collaborators included individuals from many Caribbean countries, Customs and Border Protection (CBP), and the University of Florida.

The team evaluated nine pathways: human movement, airline passenger baggage, international mail, maritime traffic (fig. 33), hitchhikers (fig. 34, fig. 35), wood packaging material, forestry, propagative materials, and natural spread. The analysts rated the pest risk associated with human movement, hitchhikers, wood packaging materials, forestry, and propagative materials as very high. The pest risk associated with airline passenger baggage, mail, and natural pest spread was rated as medium. None of the pathways assessed were rated as low risk.



Figure 33. Yachts, like those shown at a marina in Puerto Rico, enter harbors without phytosanitary inspection (Photo by Heike Meissner, CPHST, Raleigh, NC).

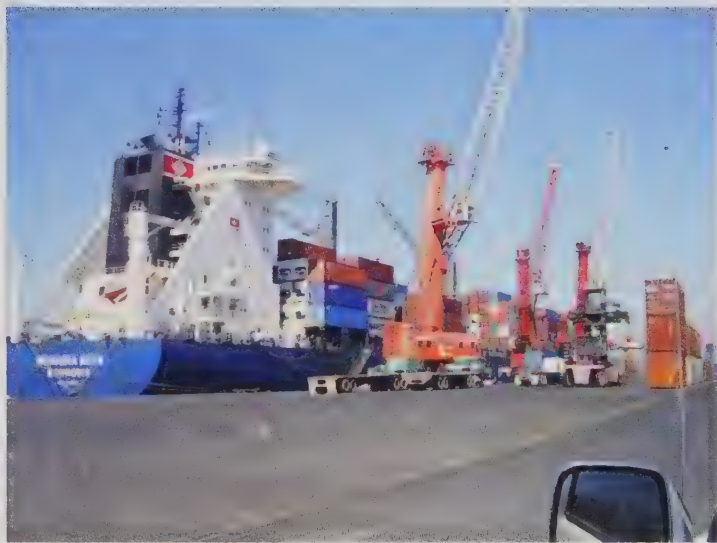


Figure 34. A cargo ship being loaded at a Caribbean port. The containers and any part of the ship may be contaminated with hitchhiker pests (Photo by Heike Meissner, CPHST, Raleigh, NC).

As part of the 250-page project report, the team listed numerous specific recommendations for improved safeguarding, many of which also have applicability outside of the GCR. The report can be accessed at <http://caribbean-doc.ncsu.edu/index.htm>.



Figure 35. These handmade craft items for sale at a Caribbean tourist market were constructed from palm leaves and present a pathway for the spread of the red palm mite (Photo by Heike Meissner, CPHST, Raleigh, NC).

Biological Control of Canada Thistle

Location: Biocontrol Unit, Raleigh, NC
Lead Scientist: Dr. Rich Hansen

Canada thistle, *Cirsium arvense* (Asteraceae) (fig. 36), is a perennial plant native to Europe and Asia that has become a widespread exotic weed in North America. Canada thistle is now found throughout most of the United States; only the far southeastern States are not generally infested. It is one of the most economically- and ecologically-damaging weeds in the United States; it occurs in a wide range of wildland, agricultural and urban habitats, and is a serious economic pest in various crops and pastures. Canada thistle reproduces vegetatively, forming dense stands that can reduce the abundance of other vegetation, including native plants. It is listed as a noxious weed in 33 States, making it the most frequently listed exotic weed in the United States.

Control of Canada thistle is usually difficult and expensive, particularly in nonagricultural habitats. Classical biological control is the release of host-specific natural enemies that feed on a weed in its native range—typically Europe or Asia—into areas where the weed has become an exotic pest. Classical biological control was recognized as a potentially useful management tool for Canada thistle beginning in the 1960s. Since then, four insect agents have been deliberately introduced in the United States. In addition, four insects and a rust fungus have been accidentally released in the United States and are occasionally employed as Canada thistle biocontrol agents. However, available biocontrol agents have either proven ineffective in controlling Canada

thistle or have been observed attacking native plants. Thus, biological control is still not a viable management tactic for Canada thistle in most of the United States.

The Canada thistle rust mite, *Aceria anthocoptes* (Acari: Eriophyidae) is native to Eastern Europe and was studied as a potential biocontrol agent for the United States beginning in the 1990s. Around 2000, *A. anthocoptes* was detected on Canada thistle plants (fig. 37) in several Eastern U.S. States. Subsequent surveys have discovered the mite in additional Eastern, Midwestern, and Western States, indicating that *A. anthocoptes* had been accidentally introduced into and established in much of the United States. Additional research was conducted to see if this immigrant mite could be exploited as a Canada thistle biocontrol agent in the United States. A key issue facing any prospective weed biocontrol agent is host-specificity—in particular, the possible impact on related



Figure 37. Canada thistle rust mites (*Aceria anthocoptes*) on Canada thistle leaf (Image by Rich Hansen, CPHST, Fort Collins, CO).



Figure 36. Canada thistle (*Cirsium arvense*) plants in Fort Collins, CO (Photo by Rich Hansen, CPHST, Fort Collins, CO).

native plants. This is particularly important for Canada thistle agents since the United States has a diverse flora of native *Cirsium* thistles (fig. 38), including more than 90 species and subspecies. Among these native plants, there are at least 14 native thistles listed as threatened or endangered. *A. anthocoptes* was believed to be specific to Canada thistle, as literature records reported it only from this plant. For this reason, the mite was considered a promising biocontrol agent.

Beginning in 2006, the CPHST Fort Collins Lab began a project to better define the host range of *A. anthocoptes* in the United States. Since previous work had demonstrated that the mite was common on Canada thistle throughout Colorado, the State's native thistle species were sampled to detect mite populations. Mites were extracted from field populations of eight native *Cirsium* species, while a California native was grown and tested in the lab. Any collected mites were submitted to taxonomists at the USDA's Systemic Entomology Lab and in Europe to determine if they were, in fact, *A. anthocoptes*.



Figure 38. Mountain thistle (*Cirsium scopulorum*) plants in Rocky Mountain National Park, CO (Photo by Rich Hansen, CPHST, Fort Collins, CO).

Through 2008, mites collected on six of the nine tested native thistles have been confirmed as *A. anthocoptes* (fig. 39). Final identification of mites from the other three native thistles is pending, but they are also believed to be *A. anthocoptes*. Thus, the mite has a broader host range than expected and appears able to feed on many native *Cirsium* thistles. For this reason, utilization of *A. anthocoptes* as a Canada thistle biocontrol agent in the United States is not recommended.

In 2009, we plan to sample additional native *Cirsium* thistles in Colorado and adjacent States for the presence of *A. anthocoptes*. Project cooperators are comparing the biology and taxonomy of U.S. and European mite populations and developing molecular techniques for more rapid identification of *Aceria* mites.



Figure 39. Canada thistle rust mite (*Aceria anthocoptes*) collected from prairie thistle (*Cirsium canescens*) in Rocky Mountain National Park, CO (Photo by G. Bauman, USDA-ARS).

Cold Treatment Program

Location: Treatment Quality Assurance Unit (TQAU), Raleigh, NC

Lead Scientist: Scott Wood

Sustained cold treatment has been employed for decades as an effective method for controlling tropical and subtropical fruit fly species. Exposing infested fruit to temperatures of 36 degrees Fahrenheit or below for a defined period results in the complete mortality of the various life stages of these insects. The procedures for utilizing this treatment method effectively, the development of treatment schedules for specific use, the approval of facility plans, the training of foreign officers to initiate treatments, and the auditing of treatment records within the PPQ Form 556 electronic database comprise a core program area of the TQAU.

Using the procedures outlined in the *USDA Treatment Manual*, arrangements are made between USDA and the National Plant Protection Organization (NPPO) of the import/export country to ship fruit using prescribed conditions. The procedures include approval of refrigeration and temperature recording equipment in International Standards Organization (ISO) shipping containers, foreign and domestic warehouses, and aboard marine vessels. Storage methods,

sensor calibration, and sensor placement are also regulated and supervised. The entire process is reviewed for accuracy upon arrival at a port-of-entry (fig. 40).



Figure 40. Container vessels transiting the Panama Canal locks. These vessels carried more than 10 million cases of cold treated fruit to U.S. ports in 2008 (Photo by TQAU staff, CPHST, Raleigh, NC).

The procedures for cold treatment are outlined in Chapter 3, “Cold Treatment,” and Chapter 6, “Certification of Cold Treatment,” of the Treatment Manual. Both of these chapters were completely revised, reformatted, and updated in 2008. The revisions include information regarding sensor placement in refrigerated containers and vessels as well as new instructions for initiating cold treatments.

Starting in 2005, the TQAU began implementation of a Web-based clearance process, the electronic 556 data system. This system is designed to expedite the clearance of cold treatment products shipped via vessels and containers (fig. 41). Over the last year, the TQAU made continued progress in upgrading and improving the electronic 556 cold treatment database. New features were added in 2008 that allow remote uploading of treatment charts by marine surveyors and allow PPQ officers to capture treatment information for all treatments, not just those initiated by foreign officers. Coupled with the built-in automated data analysis features, this allows for a completely paperless process at ports-of-entry.

In 2008, plans were reviewed and approved for refrigeration and temperature-recording equipment for three new vessels under construction in foreign shipyards. In addition, PPQ officers surveyed 69 vessels, and the TQAU reviewed all documentation. Applications for 107,000 new ISO shipping containers were approved and certified for use to conduct cold treatments of fruits. A listing of all approved and certified vessels and containers is maintained as an open-access online database and used by foreign inspectors, industry, and USDA officials to verify status before initiation of cold treatment. The database can be accessed at <https://treatments.cphst.org/vessels/>.

A major milestone was reached in 2008 when the TQAU cold treatment program achieved ISO 9001-2000 certification. This accomplishment concluded a nearly 3-year project to define and document all of our key processes and procedures, with the goal of providing better service to our stakeholders and cooperators. TQAU is now the fourth CPHST work unit or laboratory to achieve this distinction.



Figure 41. Fruit undergoing cold treatment within refrigerated containers (Photo by TQAU staff, CPHST, Raleigh, NC).



Selected Publications

Center for Plant Health Science and Technology (CPHST) employees published a number of scientific articles in 2008. The following is a selected list of those publications.

- Allan, S. A., U. R. Bernier, D. L. Kline, and M. F. Cooperband. 2008. Effects of single host odors and odor combinations on flight characteristics of *Aedes aegypti* and *Aedes albopictus*. *American Journal of Tropical Medicine and Hygiene* 79 (6 Suppl. S):1087.
- Berger, P. H., and P. J. Shiel. 2008. The beginnings of immunochemistry at the intersection of early plant virus research. *Phytopathology* 98(6):S194.
- Bock, C. H., T. R. Gottwald, A. Z. Cook, and P. E. Parker. 2008. Disadvantages of the Horsfall-Barratt scale for estimating severity of citrus canker. *Phytopathology* 98(6):S23-S24.
- Bock, C. H., P. E. Parker, A. Z. Cook, and T. R. Gottwald. 2008. Characteristics of the perception of different severity measures of citrus canker and the relationships between the various symptom types. *Plant Disease* 92(6):927-939.
- Bock, C. H., P. E. Parker, A. Z. Cook, and T. R. Gottwald. 2008. Visual rating and the use of image analysis for assessing different symptoms of citrus canker on grapefruit leaves. *Plant Disease* 92(4):530-541.
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- Chen, J., E. Civerolo, K. Tubajika, S. Livingston, and B. Higbee. 2008. Hypervariations of a protease-encoding gene, PD0218 (pspB), in *Xylella fastidiosa* strains causing almond leaf scorch and Pierce's disease in California. *Applied and Environmental Microbiology* 74(12):3652-3657.
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- Crook, D. J., L. M. Kerr, and V. C. Mastro. 2008. Distribution and fine structure of antennal sensilla in emerald ash borer (Coleoptera: Buprestidae). *Annals of the Entomological Society of America* 101(6):1103-1111.
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- Daneshgar, P., S. Jose, A. Collins, and C. Ramsey. 2008. Cogongrass (*Imperata cylindrica*), an alien invasive grass, reduces survival and productivity of an establishing pine forest. *Forest Science* 54(6):579-587.

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Appendix A. Map and List of International Partners/Collaborators

International partners and collaborators provide support to safeguarding our borders from invasive plant pests and pathogens. The Center for Plant Health Science and Technology (CPHST) collaborated in 39 countries on national and global projects (fig. 42).



Figure 42. Map of countries where CPHST worked in 2008 (*Map developed by CPHST Fort Collins Lab*).

- | | | |
|--------------------|-------------|--|
| Argentina | Guatemala | South Africa |
| Australia | Italy | Spain |
| Austria | Japan | St. Vincent and the Grenadines |
| Barbados | Kenya | Switzerland |
| Bolivia | Korea | Taiwan |
| Canada | Madagascar | Tanzania |
| Cayman Islands | Mexico | Thailand |
| China | Mozambique | Trinidad |
| Colombia | Netherlands | United Kingdom |
| Costa Rica | New Zealand | United States (including Puerto Rico and Hawaii) |
| Dominican Republic | Panama | Zambia |
| Ecuador | Peru | |
| Germany | Russia | |
| Ghana | St. Lucia | |

Appendix B. Funded Projects for Fiscal Year 2008

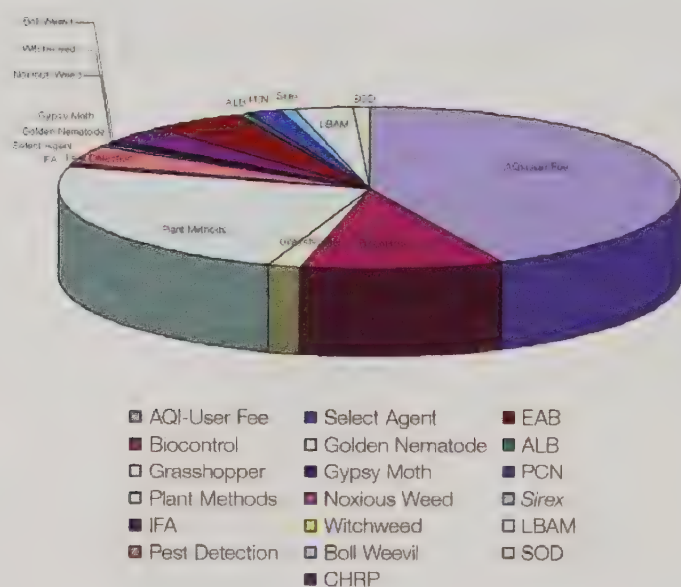


Figure 43. CPHST funding allocation in FY 2008.

Figure 43 shows a breakdown of the Center for Plant Health Science and Technology (CPHST) allocation of funding in fiscal year (FY) 2008.

Beltsville Lab

- Adapt real-time polymerase chain reaction (PCR) for the detection of the citrus variegated chlorosis strain of *Xylella fastidiosa*
- Implement PCR and real-time PCR diagnostics for rapid onsite detection of potato wart
- Improve sampling methods for detection and identification of the citrus greening bacterium
- Advance development of plum pox virus diagnostics
- Develop microelectronic chip technology for rapid detection of regulatory plant pathogens
- Adapt CANARY™ biosensors for rapid detection of regulated plant pathogens
- Develop group-specific PCR assays for the improved detection of geminiviruses and potyviruses infecting imported germplasm
- Develop and adapt real-time PCR assays for targeted viruses in foreign germplasm
- Develop lateral flow microarrays for regulated plant pathogens

- Validate PCR diagnostics methods for *Phytophthora kernoviae*
- Adapt and validate PCR detection methods for *citrus leprosis virus* (CiLV)
- Develop proficiency test reagents for high-consequence plant pathogens for delivery to NPPLAP for laboratory certification

Fort Collins Lab

Pest Identification Technologies

- Lucid Resource to Invasive Ants of the Pacific Basin
- Lucid Resource to Grasshoppers of the Western United States, Edition 3.0
- Wood Boring Beetles of the World: Lucid Resource to Wood Boring Beetle Families
- Wood Boring Beetles of the World: Lucid Resource to Genera of the *Bostrichidae*
- Wood Boring Beetles of the World: Lucid Resource to Genera of the *Buprestidae*
- Lucid Resource for Diagnosing Light Brown Apple Moth and Related Western U.S. Leafrollers (*Archipini: Tortricidae*)
- Lucid Resource to Pests and Diseases of Cultivated Palms in the United States and Caribbean Region
- Lucid Resource to Pests and Diseases of Cultivated Citrus in the United States
- Lucid Resource for the *Anastrepha daciformis*, *grandis*, *robusta*, *schausi*, and *serpentina* fruit fly species groups
- Lucid Resource to Identification of Imported Dried Botanicals
- ID Source: Creating an Identification Resource for Plant Protection and Quarantine
- RAPID-PCN: Spectral Technology for the Detection of PCN in Processed Soil Samples
- RAPID: Automation Technology Based Tools for Survey, Detection, and Identification of Pests
- RAPID-PISCES: Developing the Pest Identification Spectral Camera Experiment Station
- RAPID-RID: Developing a Remote Identification System

Biological Control of Pests

- Assess technology transfer and information services in weed management and biological control
- Assess classical biological control for management of musk thistle (*Carduus nutans*) and Scotch thistle (*Onopordum acanthium*)
- Biological control of dyer's woad (*Isatis tinctoria*) and perennial pepperweed (*Lepidium latifolium*)
- Biological control of field bindweed (*Convolvulus arvensis*)
- Biological control of garlic mustard (*Alliaria petiolata*) and yellow toadflax (*Linaria vulgaris*)
- Biological control of hoary cress (*Cardaria draba*) in the United States
- Biological control of houndstongue (*Cynoglossum officinale*)
- Biological control of orange hawkweed (*Hieracium aurantiacum*) and other invasive exotic hawkweeds
- Biological control of Russian knapweed (*Acroptilon repens*)
- Biological control of saltcedars (*Tamarix* spp.) in the Western United States
- Biological control of Canada thistle (*Cirsium arvense*)
- Survey for natural enemies of Canada thistle (*Cirsium arvense*)
- Survey for natural enemies of perennial pepperweed (*Lepidium latifolium*)
- Perfect a rearing system for light brown apple moth (*Epiphyas postvittana*)
- Develop rearing systems for root feeding biological control agents

Weed Control

- Benghal dayflower (*Commelina benghalensis* L.) control with herbicides and cover crops
- Common tansy (*Tanacetum vulgare*) control with herbicides and adjuvants: third year results
- Onionweed (*Asphodelus fistulosus*) control with herbicides and adjuvants: second year results
- Cogongrass (*Imperata cylindrica*) control with herbicides and electromagnetic backpack sprayer

- Use global positioning system (GPS) technology to estimate patch spread rate for several invasive plant species

Pest Survey and Detection

- Develop commodity-based survey documents for the CAPS community
- Develop commodity reference and survey guidelines for exotic pests of corn
- Develop commodity reference and survey guidelines for exotic pests of potato
- Conduct risk assessments for exotic pests of woody plants

Spatial Technologies

- Model pest spread based on landscape connectivity concepts
- Enhance Asian gypsy moth (AGM) (*Lymantria dispar*) trapping in the Pacific Northwest
- Model grasshopper population dynamics across the Western United States
- Conduct a spatial evaluation of how environmental variables contribute to grasshopper outbreaks
- Phenology models to predict hatch of grasshopper first instars in spring
- Evaluate global weather patterns and their relation to grasshopper populations

Gulfport Lab

The chemistry unit does not have traditional stand-alone projects, but conducts work as requested in support of other CPHST labs and APHIS programs.

Imported Fire Ant

- Biological control of the imported fire ant (IFA) using phorid flies: cooperative rearing and release program
- Biological control of the IFA: monitoring of field releases of *Thelohania solenopsae* and *Pseudacteon* spp.
- Develop new bait treatments for control of IFA
- Evaluate methods to prevent IFA from infesting baled hay and pine straw
- Develop a quarantine treatment for field grown/balled-and-burlapped (B&B) nursery stock

- Develop new treatments for containerized nursery stock
- Evaluate methods to prevent IFA from infesting commercial honey bee pollination operations
- Create a geographic information system (GIS) decision-support and management system program for monitoring and evaluating phorid fly release

Mission Lab

Fruit Fly Programs Support and Methods Development

- Evaluate augmentative release of braconid parasitoids
- Research and development support for field program activities
- Provide support for USDA-APHIS-PPQ and Florida tephritid fruit fly preventative release and eradication programs
- Evaluate tephritid fruit fly candidate bait/trap-type and portable bait stations as replacements for, or additions to detection, fly-free management, protective release, and eradication programs
- Enhance performance of male oriental fruit flies in SIT
- Supervise Guatemala fruit fly facility operations
- Assess genetically modified medfly
- Assess detection sensitivity of the California Preventive Release Program (PRP) sampling grid to *Bactrocera cucurbitae* and *B. dorsalis*
- Assess effective dose for ginger root oil (GRO)-aromatherapy at the Retalhuleu eclosion facility, Guatemala
- Conduct field assessment of combined releases of SIT and parasitoids
- Formulate Mexican fruit fly (MFF) diet
- Develop new strains of Mexican fruit flies
- Implement incubation of MFF eggs by bubbling – MFF Rearing Facility
- Assess, identify, and suppress fungal contaminants in the MFF Mass Rearing Facility
- Evaluate bait station technology (use of baits with thickeners and adhesives and non-protein attractants)
- Evaluate thermodynamics of rearing MFF

Develop Molecular Diagnostics for Arthropods and Invertebrates

- Develop molecular diagnostic techniques that identify foreign fruit fly pests
- Operate the CPHST arthropod quarantine facility
- Develop molecular techniques that identify cryptic thrip species
- Develop a molecular diagnostic tool for the genus *Anastrepha* (Tephritidae)
- Initiate the continuation of the development of molecular diagnostic techniques for mollusks of economic importance
- Transfer to Plant Health Programs a rapid molecular identification technology for immature fruit fly species
- Develop a diagnostic tool for identifying the geographic source of intercepted Dacine fruit flies (Tephritidae: Dacinae)

Biological Control and Integrated Pest Management (cross cutting)

- Develop control methods for giant African snail
- Implement biological control of tropical soda apple in east Texas using the leaf-feeding beetle *Gratiana boliviana*
- Evaluate biological control of Asian citrus psyllid in the Rio Grande Valley of Texas
- Develop classical biological control methods for Asian cycad scale (*Aulacaspis yasumatsui*)
- Develop trapping strategies to monitor for boll weevil in eradicated areas
- Support boll weevil colony
- Implement *Icerya* integrated pest management (IPM) strategies

Detect and Manage Citrus Pests and Diseases

- Survey, detect, and manage significant agricultural pests in the greater Caribbean Basin
- Monitor sentinel trees for citrus canker in Texas
- Study epidemiology of citrus canker
- Facilitate citrus canker asymptomatic study workshop
- Survey of citrus leprosis/flat mite in the Lower Rio Grande Valley, TX

- Develop management strategies for citrus greening
- Determine efficacy of packing house procedures on the mitigation of armored scales on commercial fruit

Otis Lab

Forest Pests Program Support and Methods Development

- Produce gypsy moth nuclear polyhedrosis virus
- Provide support for the Russian Far Eastern Exotic Lymantriidae Exclusion Program
- Develop and support monitoring systems for targeted exotic species
- Forecast pest potential through offshore assessments
- Assess HAACP-type methods for mitigating risk of AGM introduction
- Provide molecular analysis of male gypsy moths trapped at U.S. ports and other high-risk sites

Asian Longhorned Beetle

- Evaluate insecticides in China for the control of the Asian longhorned beetle (ALB) and other *Anoplophora* species
- Improve rearing technology for the ALB
- Investigate infestation dynamics of the ALB in North America
- Evaluate the ALB's preference of tree species and their suitability for survey and prophylactic treatments
- Apply systemic pesticides and treatment support for the ALB eradication program
- Provide molecular and genetic analyses of *Anoplophora* species

Emerald Ash Borer

- Accelerate methods development for control of exotic wood borers
- Explore emerald ash borer (EAB) biological control using exotic parasites
- Determine pesticide residue and develop assay
- Develop survey tools (traps and lures) for EAB
- Study EAB ecology: adult movement and biological control by natural enemies

- Evaluate systemic insecticides in China for the control of the EAB
- Investigate the distribution of EAB and its host trees in China
- Assess fumigation with sulfuryl fluoride as a quarantine treatment for EAB in merchantable logs and timber
- Develop dry heat, chipping, mulching, and microwave applications for use as EAB quarantine treatments
- Examine remote sensing for detection of EAB

Sirex Woodwasps

- Implement and evaluate biological control of *Sirex noctilio*
- Develop monitoring tools for *S. noctilio*
- Validate treatment options for wood commodities infested with *S. noctilio*
- Develop a degree-day model for *S. noctilio*
- Evaluate the infestation dynamics of *S. noctilio* in North America as applied to survey and management problems

Light Brown Apple Moth

- Optimize traps and lures for light brown apple moth (LBAM) detection and delimitation
- Assess and optimize mating disruption methods and materials for LBAM
- Characterize LBAM population dynamics
- Develop improved regulatory treatments for nursery stock in LBAM-infested areas
- Evaluate insecticides for control of LBAM
- Produce LBAM lures for National Survey (other than California)

Biological Control and Integrated Pest Management

- Enhance production of insect diets and life stages for use in APHIS and other cooperative research programs
- Develop automated and remotely located sensors for detecting and monitoring insect pests

Agricultural Quarantine Inspection

- Determine alternative quarantine treatments for invasive mollusks
- Evaluate quarantine treatments for bamboo
- Evaluate fumigation for control of wood-boring insects in pallet wood
- Determine Khapra beetle survey methods and diagnostics
- Evaluate radiofrequency (RF) waves as an alternative to methyl bromide for quarantine treatments
- Evaluate phosphine gas plus sustained cold treatment as a methyl bromide alternative for imported fruits
- Investigate sanitization of nematode-infested farm equipment
- Determine methods for survey, detection, and treatment of insects associated with wood packaging materials from China
- Evaluate treatments for *Bactrocera invadens* in commodities

Phoenix Lab Rearing

- Evaluate chlorhexidine gluconate (ChG) on a facility scale as an egg surface disinfectant
- Assess calco red dye concentration in artificial diet produced by the Pink Bollworm Rearing Facility
- Evaluate rearing facility moths for presence of calco red dye
- Investigate how infestation rate affects pink bollworm longevity and recapture rates in field cages
- Revise the protocols for identifying, archiving, and reviewing possible native pink bollworm moths
- Assist in an initiative to collect, identify, and catalog moths that resemble pink bollworm (look-a-like moths) in sticky traps
- Prepare and ship Singh and pink bollworm diet for LBAM production in Albany, CA, and assist in rearing and technology development for rearing LBAM
- Prepare and ship cactus moth and pink bollworm diets for cactus moth rearing in Florida and Georgia

Pink Bollworm Eradication Methods

- Acquire, prepare, and ship hundreds of pinned, labeled moths of several species, including PBW, to PPQ identifiers and other cooperators
- Test various thickening agents for effects on attractiveness and longevity of pheromone
- Substantially increase the number of documents in our PBW archive and add many unpublished documents to the archive
- Apply aerially thickened PBW mating disruption formulation to cotton fields, track first and second applications over 4 months' time of successful mating disruption under field conditions
- Finalize book chapter, "Eradication: strategies and tactics" (Chapter 23), by M.L. Walters, R. Sequeira, R. Staten, O. El-Lissy, and N. Moses Gonzales. In *Integrated Pest Management 2009*. Edward B. Radcliffe, William D. Hutchison, Rafael E. Cancelado (editors). Cambridge University Press, India.

Rangeland

- Evaluate incorporating canola oil and dry baits for improving attraction of grasshoppers and bait effectiveness on rangeland
- Replace traditional volumes of oil and water in ULV diflubenzuron sprays with alternative diluent mixes for rangeland grasshoppers
- Evaluate the manner of pick-up of diflubenzuron by *Ageneotettix deorum* and *Cordillacris occipitalis* (Orthoptera: Acrididae) from aerially sprayed rangeland
- Evaluate organic treatments for pink bollworm: evaluations of direct and indirect contact activity of spinosad and *Metarhizium anisopliae* DWR346 sprays
- Evaluate indirect contact activity of *Metarhizium anisopliae* DWR346 sprays on cotton against adult pink bollworm moths
- Evaluate initial non-target effects of *Metarhizium anisopliae* DWR346 sprays against *Lygus hesperus*
- Develop the Field Aerial Application Spray Simulation Tower Technique (FAASSTT)
- Evaluate *Metarhizium anisopliae* DWR346 dose ranges against Mormon cricket in Montana

- Further evaluate *Metarhizium anisopliae* DWR346 and new Utah Strains of *Metarhizium anisopliae* against Mormon cricket in Utah
- Increase the field activity period of spinosad against rangeland grasshoppers: Laboratory evaluation of initial mortality and residual activity of six formulations of spinosad

Plant Epidemiology and Risk Analysis Lab Commodity Pest Risk Analysis

- Produce scientific documentation in support of trade decisions regarding the importation of commodities
- Prepare pest risk assessments, identify potential mitigations, and review pest risk assessments prepared by other countries
- Identify and develop improvements in the pest risk assessment and risk management process

Exports

- Promote trade facilitation of United States agriculture exports
- Provide support to export opportunities that are blocked by technical barriers
- Prepare Export Risk Analysis Products (focus is on pest lists of arthropods and plant pathogens) associated with various commodities for export to foreign countries

Risk Analysis for Individual Organisms

- Through the New Pest Advisory Group (NPAG), assess new and imminent pest introductions into the United States and make recommendations to PPQ management regarding appropriate agency responses to exotic plant pests, including arthropods, mollusks, pathogens, and weeds

Accreditation and Certification of Risk Analysis Functions

- Through audits and improvements, maintain certification for the lab's commodity risk assessments and New Pest Advisory Group

Outreach and Training/Capacity Building/Regulatory Curricula

- Provide instructors for a regulatory science minor at North Carolina State University (NCSU)
- Engage in cooperative online courses (international standards and risk management) as part of a second regulatory curriculum program at Michigan State University
- Maintain the strong cooperative relationship established between CPHST, NCSU, and other academic institutions
- Support a regulatory curriculum that provides training to students in relevant fields on key aspects of regulatory plant protection
- Host risk analysts from other countries, pairing visiting scientists with resident analysts as mentors to provide training in risk analysis methods

Plants for Propagation (Q-37) Analyses and Regulatory Overhaul

- In support of the major initiative to revise and update 7 CFR 319.37, the quarantine that regulates the import of plants for planting, advance the regulatory process through the development of methodologies and analyses to support the APHIS decisionmaking processes associated with the evaluation of pest risk prior to authorizing the entry of propagative material into the United States

International Standards: International Plant Protection Convention (IPPC), North American Plant Protection Organization (NAPPO)

- Lend support, time, and expertise to international organizations, such as the IPPC and NAPPO, by participating on international working groups to write standards and reviewing draft standards as they become available
- Manage and maintain the Web site for the Phytosanitary Alert System (PAS) Panel, which provides oversight to early warning initiatives for NAPPO

Information Systems and Biosurveillance Analysis Forecasting

- Maintain and expand the Global Pest and Disease Database (GPDD)
- Through the NCSU/APHIS Plant Pest Forecast (NAPP-FAST) System, support the predictive pest mapping needs of the Cooperative Agricultural Pest Survey (CAPS) program and the risk assessment activities of the lab
- Generate Global Plant Hardiness Maps and post them on the NAPPFAST Web site (www.nappfast.org/)
- Create risk maps for the CAPS Top 50 Pests as well as for CAPS 2009, CAPS historical pests and CAPS commodity surveys
- Support the APHIS PPQ CAPS Program by producing pest prioritization lists using the analytic hierarchy process
- Produce and circulate the Exotic Pest Information Collection and Analysis (EPICA) notifications

Weed Risk Assessment

- Generate model and pest list for weeds
- Conduct weed analysis
- In support of the Q-37 revision, revise weed risk assessment guidelines to improve and streamline the process
- Validate the risk assessment model and compare its accuracy with that of the Australian weed risk assessment model that is being used elsewhere
- Conduct weed risk assessments of plants that pose a risk to the United States as defined by the Plant Protection Act of 2000

Caribbean Pathway Analysis

- In collaboration with the Caribbean Invasive Species Working Group (CISWG), evaluate the relative importance of the following pathways: human movement, airline passenger baggage, international mail, maritime traffic, hitchhikers, wood packaging material, forestry, propagative materials, and natural spread for the 36 greater Caribbean region (GCR) countries

- List recommendations for improved safeguarding, many of which also have applicability outside of the GCR
- Complete and circulate the resulting 250-page report entitled “Evaluation of Pathways for Exotic Plant Pest Movement into and within the Greater Caribbean Region (GCR),” posting the report at <http://caribbean-doc.ncsu.edu/index.htm>

Biological Control Unit

Since the Biocontrol Unit is comprised of a virtual team of scientists from multiple CPHST labs, biocontrol projects are listed under each lab’s project listing. One scientist, Amy Rhoda, reports directly to the CPHST Director’s Office and her projects include:

- Field release and evaluation of natural enemies of tropical soda apple
- Canal zone sentinel survey
- Offshore mitigation of invasive pests

Treatment and Quality Assurance Unit Commodity Treatment Information Systems (CTIS)

- CTIS updates for online 556 application
- CTIS implementation of online Niger Seed database application
- CTIS updates for online IRADS application

Pressure Decay Requirement for Tarpless Containerized Methyl Bromide Fumigations

- Test methods for pressure testing International Standards Organization (ISO) shipping containers
- Develop standards for pressure testing ISO shipping containers

Director’s Office Other Projects

- Develop pest diagnostics and information management

Appendix C. Abbreviations and Acronyms

AGE	Agarose Gel Electrophoresis
AGM	Asian Gypsy Moth
ALB	Asian Longhorned Beetle
AQI	Agriculture Quarantine Inspection
AQI&PT	Agriculture Quarantine Inspection and Port Technology
APHIS	Animal and Plant Health Inspection Service
ARS	Agriculture Research Service
B&B	Balled-and-Burlapped
BCU	Biological Control Unit
Bt	<i>Bacillus thuringensis</i>
CAPS	Cooperative Agricultural Pest Survey
CBP	Customs and Border Protection
ChG	Chlorhexidine Glucanate
CHRP	Citrus Health Response Program
CiLV	<i>Citrus Leprosis Virus</i>
CISWG	Caribbean Invasive Species Working Group
CPHST	Center for Plant Health Science and Technology
CTIS	Commodity Treatment Information System
DHS	U.S. Department of Homeland Security
EAB	Emerald Ash Borer
EDP	Emergency and Domestic Programs
EPICA	Exotic Pest Information Collection and Analysis
FAASSTT	Field Aerial Application Spray Simulation Tower Technique
FY	Fiscal Year
GCR	Greater Caribbean Region
GIS	Geographic Information Systems
GPDD	Global Pest and Disease Database
GRO	Ginger Root Oil
HACCP	Hazard Analysis and Critical Control Point
HLB	Huanglongbing (also known as citrus greening)
HPLC	High Pressure Liquid Chromatography
ICLN	Integrated Consortium of Laboratory Networks
IFA	Imported Fire Ant
IPM	Integrated Pest Management
IPPC	International Plant Protection Convention
IRADS	Irradiation Reporting and Accountability Database
ISO	International Standards Organization

ISPM	International Standards for Phytosanitary Measures
ITS	Internal Transcribed Spacer
LBAM	Light Brown Apple Moth
MDB	Molecular Diagnostics and Biotechnology
Medfly	Mediterranean Fruit Fly
MFF/Mexfly	Mexican Fruit Fly
NAPPEAST	NCSU/APHIS Plant Pest Forecast
NAPPO	North American Plant Protection Organization
NMSU	New Mexico State University
NPAG	New Pest Advisory Group
NPDN	National Plant Diagnostics Network
NPPLAP	National Plant Protection Laboratory Accreditation Program
PAS	Phytosanitary Alert System
PBW	Pink Boll Worm
PCN	Potato Cyst Nematode
PCR	Polymerase Chain Reaction
PERAL	Plant Epidemiology and Risk Analysis Laboratory
PISCES	Pest Identification Spectral Camera Experiment Station
PPQ	Plant Protection and Quarantine
PPV	<i>Plum Pox Virus</i>
PRA	Pest Risk Analysis
PRM	Panicle Rice Mite
PT	Proficiency Test
RAMP	Risk Analysis Mentoring Program
RAPID	Robotic Automated Pest Identification
RF	Radio Frequency
RITAWEB	Robotic Information Technology Assistant Web Interface
RPA	Risk and Pathway Analysis
RPM	Red Palm Mite
SAGARPA	Secretaría de Agricultura, Ganadería, Desarrollo Rural, Pesca y Alimentación
SDI	Survey Detection and Identification
SIT	Sterile Insect Technique
SOD	Sudden Oak Death
TQAU	Treatment Quality Assurance Unit
TSA	Tropical Soda Apple
TWG	Technical Working Group
ULV	Ultra Low Volume
USDA	United States Department of Agriculture

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